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TABLE OF CONTENTS

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INDEX

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STUDIES ON THE VEGETATIVE PROPAGATION OF PLUM ROOTSTOCKS BY LAYERING*

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A. Introduction.

Much information is now available on the influence of rootstock on scion (Hatton, 1921, 1935), the most important point being that the characteristic influence of a particular rootstock can be perpetuated by propagating it vegetatively (Hatton, 1923, Schlindler, 1932, Margolin, 1938) but is frequently lost when the rootstock is propagated from seed. As a result of the work of recent years, recommendations of world-wide application have been made for the use of particular rootstocks to suit particular conditions or scion varieties. Availability and ease of vegetative propagation are important points to be considered before a rootstock can be recommended for general use, and shortcomings in these respects have prevented several otherwise promising rootstocks from coming into commercial use.

Laurie and Chadwick (1931) have described the various methods of vegetative propagation, and intensive work on this subject has been carried out by Knight, Amos, Hatton and Witt (1926-27), Knight and Witt (1927), Stahl (1929), Tukey (1934), Hansen and Eggers (1936) and others. Experience has shown that not only different genera and species, but even closely related varieties of fruit trees frequently respond quite differently to a particular method of vegetative propagation. Hence the problem resolves itself at first into series of empirical trials with each variety. The elucidation of certain general principles is, however, of considerable value, though the application of them to each individual variety must take into account this wide variation in inherent potentialities.

Of the general principles involved, perhaps the most important is the need for good moisture conditions for successful rooting, while most fruit varieties require good aeration as well. Etiolation of the portion of stem on which root production is required is also of general value and underlies the method known as layering, distinguishing it from stooling.

Layering has been used with but little modification for the vegetative reproduction of plants for a long time. The need for raising fruit tree rootstocks vegetatively has led to the development of layering for the large scale propagation of certain varieties which do not respond favourably to the simpler stooling method. The two chief methods of layering have been fully described by Knight, Hatton, Amos and Witt (1926), Yerkes (1923), and Woodhead (1935), and their relative success depends largely on the variety concerned (Yerkes, 1926).

In the method used at East Malling, a layer bed is started by planting a row of rootstocks at a slant, about three feet apart. After one year's growth the stems of the plants are bent downwards along the row and pegged down in shallow troughs, which are then filled with a thin layer of light soil. This is done just before bud-break. The bases of the shoots that develop from the buried buds, being in darkness, become etiolated. Further soil is later banked round the bases of these shoots as they become longer, so that the underground portion of each may eventually be considerably longer than the part originally etiolated. At the end of the season, the layers are unearthed and the shoots that have rooted are cut off and planted out as rootstocks. Unrooted shoots are also sometimes cut off, if well etiolated, and may root when planted out as cuttings. Normally the more vigorous shoots, and those which have

* Investigations carried out at East Malling Research Station, Kent, England, between 1939-41, during a period of post-graduate study. The present paper is part of a Thesis approved for the Degree of Doctor of Philosophy in the University of London.

failed to root, are pegged down flat alongside the original layer. If every shoot has rooted care is taken to leave a sufficient number of them for further rootstock production.

The economic value of the method depends mainly on the number of rooted shoots obtained per yard run of the layer bed, and this depends on the number of the shoots produced and the percentage of them that develop roots. Varieties differ widely in these respects, and unfortunately some of the most useful rootstocks tend to produce very few rooted shoots. Rooting varies, however, from season to season, and different degrees of rooting may occur in a single season on different parts of the same layered plant. This suggests that rooting depends on a number of internal and external factors, some of which may be controllable, and that fuller knowledge of these factors might lead to improvements in cultural practice and more successful rooting.

With this in mind, a census of rooting and of shoot growth in a plum layer bed was carried out in the autumn of 1938 and repeated in 1939. Data from a similar census, carried out by S. R. Gandhi in 1932, were also utilized. The material selected was Pershore (Yellow Egg), a shy rooting but valuable rootstock. The results of the three censuses are described in Section B. Certain modifications in cultural treatment suggested by these results were tried out in 1940, and they are described in Section C. Finally, synthetic growth-substances have recently been applied to cuttings and to intact plants for the stimulation of root production with some success (Cooper (1935-38), Laibach (1935), Tincker (1936, 1938, 1940), and Pearse (1939*a*)), and Pearse (1939*b*) has fully reviewed the voluminous literature on this subject. Hence, the application of a growth-substance to the layers and to the shoots arising from them was tried out, and the results are also described in Section C.

B. OBSERVATIONS ON LAYER BEDS OF PERSHORE PLUM.

The observations were carried out during the autumn months, and were made on (i) the parent plant and (ii) the one-year-old shoots arising from the layers. The number of parent plants examined, the number of layers per plant and the lengths of the layers were recorded. The number of roots, the lengths of the shoots, the length of the etiolated portion of each, the distance of the shoot from the base of the parent plant, its position on the layer, its diameter and type of terminal growth were also recorded. Certain measurements made in 1932 could not be repeated in later years on the older plants, but a few additional measurements were included in 1938 and 1939.

Effect of season and age of layer beds on shoot and root production.

As the layer beds were of different ages in the three seasons, it is impossible to separate for certain the effects of age from those of season. The total number of shoots, and the percentage that rooted are shown in Table I. χ^2 tests (Fisher, 1934) were carried out on the

Table I.

The effect of season and age of layer bed on shoot and root production.

	1932.	1938.	1939.
Age of layer beds	2 years	8 years	9 years.
Number of parent plants	100	98	65
Total number of shoots	599	1,062	854
Percentage rooted	59	36	42
Total rainfall	21·5 in.	26·8 in.	33·0 in.
Rainfall for May, June, and July ..	5·9 in.	4·2 in.	5·5 in.

numbers rooted and non-rooted, and rooting was found to be significantly greater in 1932 ; there was no significant difference between the values for 1938 and 1939. The higher percentage in 1932

may have been due to the parent plants having been younger ; they were then only two years old whereas in 1938 and 1939 they were 8 and 9 years old respectively. In this connection it is of interest to note that the number of shoots per parent was significantly less in 1932 than in the two later years ; the greater number of shoots in the later years may have resulted in greater competition in the struggle for existence and this, in its turn, to less successful rooting, although past experience has suggested that rooting performance, in many types of rootstocks, tends to improve with increasing age (Knight, Amos, Hatton and Witt, 1926-27). Another possibility might be seasonable differences in rainfall in the three years, and the values for these are included in Table I, but examination of them showed that there was no connection between them and rooting performance. It is possible that the rainfall at the critical time for growth, May, June and July, may have influenced rooting, and the relevant figures are also included in Table I.

THE EFFECT OF NUMBER OF LAYERS PER PARENT PLANT ON SHOOT AND ROOT PRODUCTION.

In the first year the parent plants have one layer each, but in later years the number varies as new shoots are pegged down and old ones are removed or rot away. The parent plants were grouped according to the number of layers borne by each, and a statistical analysis was carried out to determine how far this might have influenced the percentage of shoots that rooted. The results indicated that there was no significant correlation between the number of layers and rooting performance of the shoots in any season.

THE EFFECT OF LENGTH OF LAYER ON SHOOT AND ROOT PRODUCTION.

In the 1932 census, the length of the layers was measured to the nearest centimetre. For convenience in analysis the layers have been arranged in groups according to their lengths, as shown in Table II. It would appear that the shorter the layers the higher the percentage

TABLE II.

Relation of length of layer to shoot and root production.

Length of layer.	Up to 60 cm.	61-120 cm.	121-180 cm.	Over 181 cm.
Number of layers*	19	68	55	2
Total shoots	44	277	254	13
Percentage rooted	77·2	59·9	55·1	38·4

* Lengths of four layers, bearing 11 shoots, not recorded.

of shoots that rooted (P = ·01). Layers from 61 to 120 cm. long had nearly double as many shoots as those less than 61 cm. long. However, greater length of layer was not accompanied by a proportional increase in the number of shoots per foot run of layer. The inference is that there is no advantage in using long layers.

Owing to practical difficulties, arising from the more complicated nature of the layer system in older layer-beds, the total length of individual layers could not be measured in 1938 and 1939.

THE EFFECT OF DISTANCE OF SHOOT FROM BASE OF PARENT PLANT ON SHOOT AND ROOT PRODUCTION.

The distance of the point of origin of each shoot from the base of the parent plant was measured to the nearest centimetre at each census. For convenience in statistical analysis the shoots were arranged in groups as shown in Table III. The analysis was carried out by the χ^2 method on the numbers of rooted and non-rooted shoots. The values shown in the Table indicate that the distance of a shoot from the base of the parent plant had a considerable effect on its rooting performance, but that the effect was in opposite directions in 1932 and in 1938 and 1939. In 1932 the percentage that rooted increased with increasing distance, whilst in the latter two years the shoots nearest the base rooted best. The differences were significant

TABLE III.

Effect of distance from parent base on shoot and root production.

Distance in cm.	1932.*		1938.†		1939.†	
	No. of shoots.	% rooted.	No. of shoots.	% rooted.	No. of shoots.	% rooted.
0- 60 cm.	368	57·3	434	47·2	514	43·5
61-120 cm.	210	58·5	433	27·7	269	39·8
121-180 cm.	20	85·5	172	22·6	53	33·9
181-240 cm.	—	—	21	33·3	16	56·2
	$P < \cdot 05$		$P < \cdot 01$		n.s.	

* Shoots more than 180 cm. from base omitted. † Shoots more than 240 cm. from base omitted.

in 1932 and 1938, but not in 1939. The most obvious reason for these differences would seem to be the age of the layer beds. In 1932 they were only two years old, while in 1938 and 1939 they were 8 and 9 years old.

At first sight the values for 1932 given in Table III seem inconsistent with those in Table II, for in the one the more distant shoots root best, while in the other the shoots on the longer layers rooted worst. The reason for this apparent discrepancy is made clear by Table IV. Here the shoots have been grouped into three classes :

(i) those on layers less than 121 cm. in length,
(ii) those less than 121 cm. from base of parent plant on layers more than 120 cm. in length.
(iii) those more than 120 cm. from base on layers more than 120 cm. in length.

TABLE IV.

Rooting at different distances from base on layers of different lengths.

Length in cm.	Distance in cm.			
	0-120		Over 120	
	Total shoots.	% rooted.	Total shoots.	% rooted.
0-120	321	62·3		
Over 120	246	53·6	21	80·9

The highest percentage of rooting occurred in the last class, but the number of shoots concerned was small; rooting nearer the parent plant was worse on the long than on the short layers, and this more than counterbalanced the higher rate among the few shoots at the greater distances. In general, therefore, the results seem to suggest that it is a disadvantage to leave the layers long.

THE EFFECT OF DISTANCE OF SHOOT FROM BASE OF PARENT PLANT ON DIAMETER OF SHOOT.

The diameter of each individual shoot was measured with a slotted gauge at a point 5 cm. above its base. Shoots that would enter the 3 mm. slot but were too large for the 2 mm. slot were denoted 3 mm., those that would enter the 4 mm. slot but were too large for the 3 mm. were denoted 4 mm., and so forth. The largest slot was 15 mm. and the few shoots too large for this were denoted " over 15 mm." Shoots under 2 mm. were ignored. As the exact diameters of the shoots over 15 mm. were not measured it was not possible to calculate the mean diameter at different distances from the base, but there seemed to be a tendency for the diameter to decrease with increasing distance up to a point ; beyond this point distance seemed to have little effect on diameter.

THE EFFECT OF DISTANCE OF SHOOT FROM BASE OF PARENT PLANT ON LENGTH OF SHOOT.

The lengths of the shoots were determined in 1932 alone, for in 1938 and 1939 the tips of some of them had been removed to make cuttings.

In Table V the shoots have been grouped according to the distance of their point of origin from the base of the parent plant ; the number in each group and the mean lengths are given. It will be seen that there is a general tendency for the lengths to decrease as the distance from the base increases.

TABLE V.

Effect of distance of shoot from base of parent on length of shoot.

Distance (cm.)	Total shoots.	Average lengths (cm.)
0-60	368	101·7
61-120	210	75·6
121-180	20	62·5

RELATION BETWEEN DIAMETER OF SHOOT AND ROOTING PERFORMANCE.

Although the diameter of the shoot cannot directly be controlled, it may be influenced indirectly through its connection with some factor, such as length of layer, which can be controlled. A knowledge of the relative rooting capacity of shoots of different diameter is therefore of some practical importance, apart from its theoretical interest. For convenience in analysis the shoots were grouped as shown in Table VI. To economize space here, only the total number of shoots and the percentages that rooted are shown, but the analysis was carried out by the χ^2 method on the numbers rooted and non-rooted. The levels of significance are shown in the Table. In 1938 and 1939 there was a strong tendency for rooting performance to improve with increasing diameter, especially amongst the thinner shoots. There was some indication of a similar tendency amongst the thinnest shoots in 1932, but in that season the shoots with the greatest diameter rooted worst. It is possible, of course, that the greater diameters were due to better rooting and not vice versa ; for a shoot well-established on its own roots might grow larger than one dependent on the more distant roots of its parent plant.

TABLE VI.

Relation of shoot diameter to root production.

Diameter in cm.	1932.		1938.		1939.	
	Total shoots.	% rooted.	Total shoots.	% rooted.	Total shoots.	% rooted.
3, 4, 5	233	55·7	437	21·0	344	34·6
6, 7, 8	145	65·5	263	33·4	253	39·5
9, 10, 11	100	60·0	164	47·5	97	53·6
12, 13, 14	75	60·0	95	54·7	68	50·0
15 and over	46	43·4	103	60·1	92	56·5
	$P = \cdot 05$		$P < \cdot 01$		$P < \cdot 01$	

THE EFFECT OF LENGTH OF SHOOT AND OF ITS ETIOLATED PORTION ON ROOTING.

Total length of shoot is another character not under direct control. The data for 1932 were examined statistically, but no significant difference in rooting amongst shoots of different lengths was found.

The length of the etiolated portion was determined for some of the shoots in 1932, but could not be repeated in the later censuses. A random sample of 36 of these shoots was taken from the data for purposes of analysis, and the correlation coefficient between the length and the number of roots determined. The value of the coefficient was found to be $+ \cdot 474$ ($P < \cdot 01$).

Relation of position of bud on layer to shoot and root production.

The buds are arranged spirally on the shoots. When the latter are pegged down horizontally as layers, some of the buds are on the lower surface, others on the upper, and the remainder more or less at the sides. Shoots arising from buds on the lower surface grow almost in a semicircle before they come up vertically through the soil. Those developing from buds at the sides often run parallel to the layer for some distance before turning up. Shoots from the buds on the upper surface are quite straight and vertical.

When recording the rooting performance of individual shoots in 1938 and 1939, the position of the bud from which each arose was noted. The results are summarized in Table VII, in which the terms top, side and bottom refer to shoots arising from buds on the upper surface,

Table VII.

Relation of location of bud on layer to shoot and root production.

	1938.		1939.	
	Total shoots.	% rooted.	Total shoots.	% rooted.
Top	504	26·5	433	35·3
Side	439	39·4	342	45·3
Bottom	119	54·6	79	62·0

side position and lower surface of the layer, respectively. Shoots from bottom buds rooted best, those from top worst. Statistical analysis showed that the difference between the value for the top shoots and those for the bottom and side shoots taken together was highly significant, whilst that between bottom and side shoots was not significant. As already mentioned, bottom and side shoots curve or bend below ground, this may be why they root better than top shoots. Another likely reason is that the etiolated portion of each is longer than that of the straight vertical shoots arising from top buds. Whatever the reason, the results suggest a possible method for improving rooting by stimulating more side and bottom buds to break.

The effect of type of shoot growth : checked or unchecked.

In certain kinds of plum rootstock, including some which root readily like Myrobolan B., the main stem ceases growth early and the shoot growth is continued by laterals arising from axillary buds. In other kinds, including the shy-rooting Pershore, the terminal bud of the main stem remains active until later in the season, and the growth of the laterals is subordinate to that of the main stem. Occasionally in Pershore, the main stem may stop growth early, and laterals may push out. This is usually due to damage to the terminal growing point by insects or other causes. The behaviour of individual shoots in this respect can readily be determined in most cases by examination during the dormant period. Where terminal growth has ceased early, laterals are usually longer and are grouped near the apex, the longest tending to be near the tip. Where terminal growth has continued throughout the season, such laterals as are present tend to be smaller and lower down on the main stem.

At the 1938 and 1939 censuses each shoot was marked " S " or " N " according to whether or not the growth of its main stem appeared to have stopped early in the season. The total number of shoots and the percentage that rooted in each group are shown in Table VIII.

TABLE VIII.

Effect of checking shoot growth on shoot and root production.

	1938.		1939.	
	Total shoots.	% rooted.	Total shoots.	% rooted.
Checked	410	21·0	103	34·0
Non-checked	652	43·9	751	42·9
	P < ·01		n.s.	

Percentage rooting was apparently influenced by the type of top growth of the shoots, a higher percentage being obtained when the tips of the shoots continued growth until late in the season. The difference was thoroughly significant in 1938, but not in 1939. Thus, early cessation of growth of the main stem in Pershore seems to reduce rather than increase rooting.

C. EXPERIMENTAL.

The observations made on the layer beds indicated that shoots arising at a distance less than 120 cm. from the base of the parent plant root better than those farther away, and that those arising from buds situated at the sides or on the lower surface of the layers root better than those from buds on the upper surface. It would seem therefore that rooting might be improved by shortening the layers and by rubbing off the buds on the upper surface. Such treatment might force dormant buds on the layers to break in positions where the resulting shoots would be likely to root. It would also seem possible that rooting might be improved by treating the layers with a growth-promoting substance. Experiments carried out to test these possibilities are described below.

THE EFFECT OF SHORTENING LAYERS ON SHOOT AND ROOT PRODUCTION.

Shoots were cut back to lengths of 120 cm., 90 cm., and 60 cm. before they were pegged down as layers. For control purposes some of the shoots were left unshortened. The experiment was laid out in 12 blocks of four plants each, three plants having their shoots shortened and the fourth left unshortened. The four treatments were randomized in each block and an attempt was made to choose plants as uniform as possible.

At first sight it might seem likely that shortening the layers would reduce the total number of shoots per plant, and that this might cancel out any gain from improved rooting. The results of the treatments on the total number of shoots as well as on their rooting performance were therefore examined statistically. For rooting performance the χ^2 method (Fisher 1934) was used, and for number of shoots an analysis of variance was carried out, the numbers being first transformed into square roots ($\sqrt{n + \frac{1}{2}}$) as recommended by Bartlett (1936). These transformed values are given in italics in Tables IX and XI. The variance due to treatments was split into single degrees of freedom and each compared with its appropriate error.

TABLE IX.

The effect of shortening the layers on shoot and root production.

Treatment.	Shoots on 12 plants.		% rooted.*	Shoots per plant.	
	Rooted.	Non-rooted.		Total.	$\sqrt{n+\frac{1}{2}}$*
Control	36	99	26·6	11·3	*3·28*
120 cm.	43	91	32·1	11·2	*3·29*
90 cm.	53	93	36·3	12·2	*3·40*
60 cm.	63	101	38·4	13·7	*3·63*

* Treatments showing non-significant differences bracketed together.

As shown in Table IX, shortening the layers, even to 60 cm., did not reduce the number of shoots per plant; indeed, the number of shoots actually increased with the severity of shortening, but not significantly. It seems that shortening the layers forced into growth buds that would normally have remained dormant and that this more than compensated for those cut off. Rooting performance, likewise, improved with severity of shortening. Taken as a whole, the differences between the values for the four treatments were not significant, but further χ^2 tests by Yates's (1934) method, carried out between each pair of treatments, showed that layers shortened to 60 cm. had a significantly higher proportion of rooted shoots than the unshortened controls. The number of shoots per 12 plants was increased from 135 to 164 and the percentage rooting from 26·6 to 38·4.

THE RELATION OF POSITION OF THE BUDS ON THE LAYER TO ROOT PRODUCTION.

The rooting of shoots from buds in different positions was again examined. For the χ^2 test, shoots from side and bottom buds were grouped together, because it had previously been shown that those from these two positions did not differ significantly in rooting performance. The figures are given in Table X, and they show that shoots from the side and bottom buds rooted significantly better than those from top buds (P = ·01). This confirms the results already obtained in the censuses of the layer beds.

TABLE X.

Relation of position of buds to shoot and root production.

Position.	Shoots on 48 plants.		% rooted.
	Rooted.	Non-rooted.	
Top	60	213	22·0
Side	108 } 135	139 } 171	44·1
Bottom	27 }	32 }	

THE EFFECT OF PARTIAL DISBUDDING OF THE LAYERS ON SHOOT AND ROOT PRODUCTION.

The main purpose of this experiment was to see if the number of rooted shoots per plant could be increased by rubbing out incipient shoots developing from top buds, at the time of bud break, thereby stimulating more side and bottom buds to break dormancy. To compare the effect of rubbing out, as such, with that of the position of the buds rubbed out, a further treatment was introduced in which bottom buds, instead of top buds, were rubbed out. The treatments were carried out just before the layers were pegged down and covered with earth. The experiment was laid out in 15 blocks of three plants each, one plant under each treatment with one untreated as a control. The treatments were randomized in each block and care was taken to select plants as uniform as possible. The significance of differences in rooting performance and in numbers of shoots per plant were examined statistically, as before, by the χ^2 method and analysis of variance respectively. The results are given in Table XI.

TABLE XI.

The effect of partial disbudding on shoot and root production.

Treatment.	Shoots on 15 plants.		% rooted.*	Shoots per plant.*			
				Total.		Rooted.	
	Rooted.	Non-rooted.		No.	$\sqrt{n+\frac{1}{2}}$	No.	$\sqrt{n+\frac{1}{2}}$
Control	36	92	28·1	8·53	2·92 }	2·40	1·59
Top buds off	63	83	43·2 }	9·73	3·03 }	4·20	2·05 }
Bottom buds off	56	80	41·2 }	9·07	2·91 }	3·73	1·92 }

* Non-significant differences (P > ·05) are bracketed together.

It will be seen that partial disbudding significantly improved rooting performance, whether the top or the bottom buds were removed. The position of the buds removed had no significant effect. The number of rooted shoots per plant was significantly greater on disbudded plants ; the total number of shoots was also greater, but not significantly so.

Treatment of layers and shoots with a growth-substance.

Five concentrations of indole-butyric acid (0·0, 0·25, 0·5, 1·0 and 2·0 per cent.) were prepared in lanoline (Laibach, 1935), and applied to the layers and shoots of Myrobolan B., Pershore and Pear C8, at four different times during the growing period, viz. (i) at bud break, (ii) when the shoots were from 1 to 2 inches high, (iii) when the shoots were 6 to 8 inches high, and (iv) when the shoots were full grown.

For the first application small incisions were made under the bark of the parent layers, close to the growing buds, and a small amount of the paste was inserted. The layers were then earthed up. At the second application the shoots were too tender for incisions to be made in them without injury ; the pastes were therefore smeared round their bases. At the third and fourth applications the shoots were strong enough to bear the shock of incisions ; small cuts were therefore made at the bases of the shoots and the lanolin paste was inserted. For the last three applications the layers had to be unearthed and then earthed up again.

There was no apparent effect on the poor rooting subjects, Pershore and Pear C8, neither was there any response to the first and second applications, even on the normally ready-rooting Myrobolan B. The third and fourth applications, however, had a definite influence on the amount and type of rooting of this variety. The place of application became swollen with an enormous mass of callus, and bunches of roots were noticed, mainly on the upper edges of the incisions. No swellings occurred in the control plants.

D. GENERAL CONCLUSIONS.

The success of layering as a method of vegetative propagation depends largely on the number of rooted shoots obtained per foot run of layer bed, and this in its turn depends on the total number of shoots and the percentage of these that root. Any cultural treatment that leads to an increase in the number of rooted shoots per foot run will be of economic value, provided it does not involve a proportional increase in the cost of cultivation. Observations of the rooting performance in layer beds of the plum rootstock Pershore, showed that it was best on short layers, in regions near the parent plant, and on shoots arising from buds at the sides and on the undersurface of the layers. These observations suggested that rooting might be improved by shortening the length of the layers and by rubbing out the top buds, but there was a danger that these practices might reduce the total number of shoots. The results of the experiments carried out showed that these two operations did improve rooting without reducing the total number of shoots ; in fact, a slight increase in their number was noticed. The following recommendations can therefore be made for this rootstock :

(i) Shoots before layering should be cut back to about 2 feet. When setting up a new layer-bed the plants should be spaced about 18 inches apart, instead of 3 feet as at present practised. In old-established beds, new plants might be inserted between the old ones, but it is doubtful if plants inserted thus would make good growth.

(ii) A proportion of the buds should be removed, and as far as possible those from the upper surface should be chosen for removal.

These two recommendations are based on the results obtained with only one variety, Pershore, under East Malling conditions. Before they can be put forward as of general application they should be tested on other varieties and in other localities. Moreover, the operations

were carried out at the time of earthing up, at bud-break. Other times in the dormant season might be tried and the results compared.

The application of a growth-substance to layers and shoots did not result in any improvement in rooting performance of plants difficult to root, whilst with an easily rooting variety, such as Myrobolan B., an improvement was noticed only when application was made at a time which involved unearthing and re-earthing the layers. These processes involve much trouble and expense as well as considerable risk to the plants, especially in dry weather. Hence this treatment cannot be recommended until improvements in the method of application can be made.

SUMMARY.

Censuses of rooting and of shoot growth in Pershore plum layer beds carried out during the autumns of 1932, 1938 and 1939, showed that :

1. Rooting varied from year to year, and there was some indication that it was influenced by the rainfall during the critical period for growth.

2. The number of layers per plant had no significant influence on the rooting of the shoots.

3. Short layers had proportionately more shoots than long ones, and the percentage rooting progressively decreased with increasing length of layer.

4. The number of shoots and the percentage of them that rooted were significantly greater when they were situated near the base of the parent plant than when farther away.

5. The diameter of the shoots showed a tendency to decrease with increasing distance from the parent plant.

6. Percentage rooting increased with increasing shoot diameter up to a certain point ; beyond this no clear conclusions could be drawn.

7. Shoots nearer the base of the parent plant were longer than those farther away.

8. No correlation was found between length of shoot and rooting performance.

9. The longer the etiolated portion of the shoot, the larger the number of roots.

10. Shoots arising from buds on the under-surface and the sides of the layers rooted better than those from buds on the upper surface.

11. Shoots that continued terminal growth until late into the season rooted significantly better than those which ceased growth earlier.

12. Experiments carried out on the layer beds to improve rooting showed that it could be significantly increased, without reduction in the number of shoots, by :

(*a*) Cutting back the layers to about 2 feet.

(*b*) Partial disbudding of the shoots at the time of layering.

13. Application of lanolin paste containing indole-butyric acid in various concentrations did not affect rooting in Pershore plum or Pear C8 layers ; but rooting in Myrobolan B was improved by summer application to the shoots. Most of the roots appeared at the point of application, where a large callus developed.

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THE INCIDENCE OF SUPERFICIAL SCALDS IN APPLES GROWN IN SOUTH AFRICA IN RELATION TO STORAGE TEMPERATURES

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LITTLE information is available from earlier work as to the connection between superficial scald in stored apples and storage temperature. Emphasis in the past has mainly been laid on the effects of ventilation, still air, pre-storage delay and the use of oiled wrappers, the tests often having been made at one temperature only. In the work to be described here the chief aim was to determine the effect of different storage temperatures on the occurrence of skin discoloration covered by the general term Superficial Scald, but the effect of oiled wrappers has also been studied though not that of pre-storage treatment.

The existence of two well marked types of superficial scald has been detected, with the possibility of a third. It is proposed to designate these two (*a*) *Superficial Scald*, developed only at temperatures ranging from 1·7° C. (at which it is only slight) to 7·2° C., and (*b*) *Frigescence Superficial Scald*,* developing most typically at temperatures between −1·7° C. and 1·1° C. The further characteristics of the two types will be dealt with in detail later.

The storage tests were carried out during four seasons, with fruit obtained mainly from the S.W. Cape Province, but also from the Langkloof, in the E. Cape Province, and from Vereeniging in the Transvaal. The varieties chiefly employed were: Ohenimuri, Red Delicious, Granny Smith and Rome Beauty, but Wemmershoek and White Winter Pearmain were used to some extent.

At the time of storage the fruits were at the stage of ripeness usual for export apples. They were stored in the dark, under conditions of rapid and continuous air circulation in chambers at specific constant temperatures ranging from −1·7° C. to 7·2° C. After withdrawal from store they were unwrapped, examined, laid in single layers on open trays and transferred to a dark room at 18·3° C. or 20° C., where they remained for one or two weeks under continuous air circulation. Often little or no scald was apparent immediately on removal from store, but it became evident after transference to the higher temperature; little or no further scald developed during a second week's retention at 18·3° C. or 20° C.

The scalds encountered varied considerably in appearance. One form consisted mainly of more or less circular areas surrounding lenticels, of diameters varying from less than 1 to more than 5 mm., intermediate ones being most frequent. Another form appeared as clear areas centred on lenticels and surrounded by scalded skin; in others the scald was of a diffuse character, with or without slight depression of affected areas. In recording the colours of the scalds Séguy's "Code Universal des Couleurs" was used (20). They varied from dirty grey (*405*) or green (*207-8, 223, 327-8, 474-5*) through light (*217-19, 261-3, 339*) to medium brown (*336-8*) to dark brown (*176, 701*), these designations, however, being indicative rather than strictly precise.†

The unit of fruit examined for each storage temperature and treatment (oiled or plain wrappers) was a bushel box, usually containing 125 or 150 apples. Unless otherwise stated,

* Term adopted after consultation with Prof. R. S. Adamson.

† A "spotting" type of superficial scald has been distinguished by Kidd and West (13) and recorded for Rome Beauty by Isaac and Boyes (10, 11).

the values assigned for superficial scald are for apples kept at 18·3° C. or 20° C. for one week after withdrawal from store. In making counts, apples showing storage pit were excluded; this disorder was serious only in the variety Red Delicious.* Correction was made for fruits discarded on removal from store mainly because of fungus attack. The amount of superficial scald arising is expressed by the terms (*a*) prevalence and (*b*) intensity. The former denotes the number of apples affected and is the criterion mostly used; the latter indicates the degree of attack on individual apples and implies not only the area of the scalded surface, but also its appearance, such as lighter or darker brown discoloration and whether the surface was slightly sunken or not. No attempt has been made to express intensity quantitatively. Prevalence is used in the same sense as that employed by Kidd and West (14), but intensity, as used by these investigators, denoted the amount of scalded surface shown by individual apples.

Superficial Scald in Ohenimuri Apples.

Storage tests were made with fruit from the same farm at Elgin in 1939 and 1940, in the former year at −1·7°, 0°, 1·7° and 3·3° C., and, in the latter, at 7·2° C. in addition. The values for superficial scald obtained in the two years for the first range of temperatures are plotted in Fig. 1, A and B. It is clear that it developed only to a limited extent at −1·7° and 0° C., and was associated with the higher temperatures, 1·7° C. and 3·3° C., being more prevalent

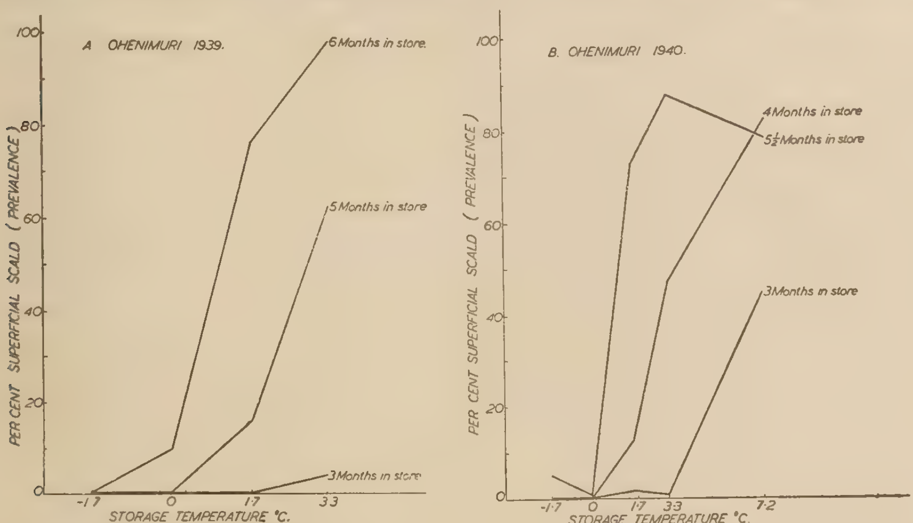

FIG. 1.
Superficial scald (prevalence) of Ohenimuri apples in relation to the temperature and length of storage.

at the latter than the former; with increased temperature there was also increased intensity of scald. During the first three months of storage in both years, the prevalence was less than 5 per cent., except at 7·2° C.; but during the fifth and sixth months in 1939 there was rapid increase at 1·7° C. and at 3·3° C. In 1940 the scald developed rapidly at 3·3° C. during the fourth month of storage, but at 1·7° C. this increase occurred during the fifth month. The figures for 7·2° C. are given in Table I, from which it is clear that while the scald was well developed by the end of the third month at 7·2° C., its prevalence after five and a half months at this temperature was somewhat less than at 3·3° C. The intensity also, was less at the higher temperature. If it is true that this scale is associated with the formation of esters and acetaldehyde, its decrease at 7·2° C. may be due to the increased volatility of these substances at the higher temperature.

* It is intended to deal with the relationships between superficial scald and storage pit in a separate paper.

TABLE I.

Per cent. superficial scald of Ohenimuri apples stored at 7·2° C. (1940).

Months in store (+1 week at 20° C.)	Superficial scald per cent.	Superficial scald at 3·3° C. for comparison.
3	44·4	0·7
4	82·0	47·0
5½	78·6	87·5

In 1939 oiled wrappers had little or no effect on prevalence and this was confirmed in 1940, the data for that year being given in Table II. These wrappers, however, modified the character

TABLE II.

Effect of oiled wrappers on the prevalence of superficial scald in Ohenimuri apples (1940).

Months in store (+1 week at 20° C.)	Storage temperature ° C.	Per cent. superficial scald.	
		Plain wrappers.	Oiled wrappers.
3	7·2	44·4	38·2
4	3·3	47·0	30·4
5½	−1·7	4·3	0·0
	0·0	0·7	2·0
	1·7	73·0	69·6
	3·3	87·5	77·0
	7·2	78·6	82·0

of the scald, for fruits in sulphite wrappers showed a widely diffused, light brown (*217-19, 339, 261-3*) type of it, whereas those in oiled wrappers showed a smaller area of scalded surface and the

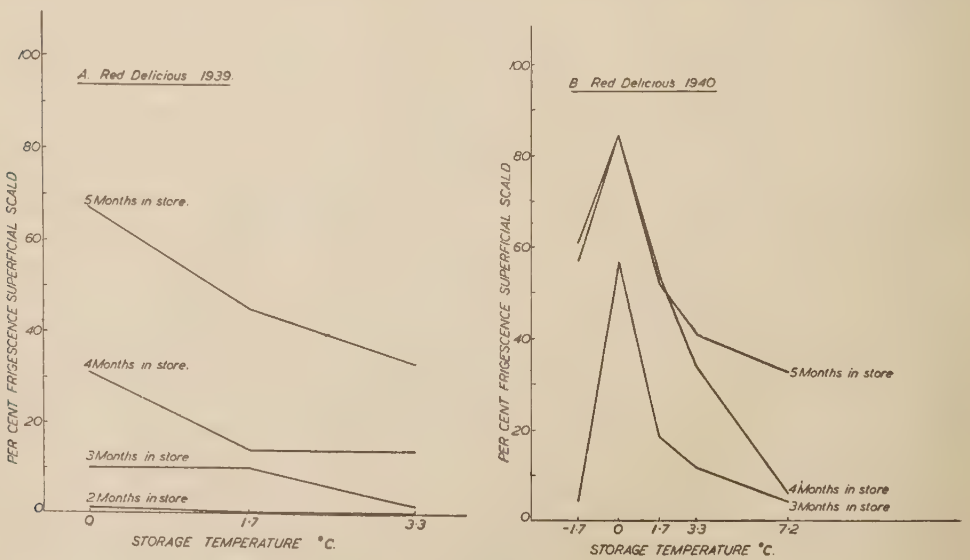

FIG. 2.

Frigescence superficial scald (prevalence) of Red Delicious apples in relation to the temperature and length of storage.

colour was greyish-green (*207-8, 327-8*) or very light brown, the fruit being on the whole more normal and attractive in appearance.

FRIGESCENCE SUPERFICIAL SCALD IN RED DELICIOUS APPLES.

Storage tests were made with fruit of this variety from the same farm at Elgin in 1939 and 1940 at 0° C., 1·7° C. and 3·3° C. and at −1·7° C. and 7·2° C., in addition, in the latter year. The effect of temperature on the development of scald is illustrated in Fig. 2, A and B, from which it will be seen that it was of the frigescence type, developing pre-eminently at 0° C. and decreasing decidedly with increased storage temperature. At and after the end of the fourth month a considerable amount developed at −1·7° C., but it was always less at this temperature than at 0° C. Its intensity followed its prevalence and was greatest at 0° C. As compared with that in Ohenimuri apples the scald at its temperature of optimum development was more prevalent earlier during storage.

Storage tests with oiled wrappers in 1940 showed that whilst they did not completely control this scald they considerably reduced its prevalence and intensity, and its essential character was the same whether plain sulphite or oiled wrappers were used. The relevant figures are given in Table III.

TABLE III.

Effect of oiled wrappers on the development of frigescence superficial scald in Red Delicious apples (1940).

Months in store (+1 week at 20° C.)	Storage temperature ° C.	Per cent. frigescence superficial scald.	
		Plain wrappers.	Oiled wrappers.
3	−1·7	3·8	3·8
	0·0	57·0	27·2
	1·7	18·8	4·4
	3·3	11·4	2·5
	7·2	4·3	0·0
4	−1·7	61·0	33·3
	0·0	84·2	48·5
	1·7	53·0	19·4
	3·3	33·6	1·0
	7·2	5·8	0·0
5*	−1·7	57·0	16·3
	0·0	84·0	—
	1·7	52·2	12·4
	3·3	41·0	5·3
	7·2	32·7	5·4

* Apples kept at 20° C. for 9 days after removal from store.

The scald was medium brown (*338-336*)* in colour, diffused over the surface and unsunken or only slightly sunken. After four months storage at 7·2° C. the fruit was rather dull in colour, this being rather less marked in the apples in oiled wrappers. After five months, this dullness was more marked in the fruit in both plain and oiled wrappers.

THE SUPERFICIAL SCALDS OF GRANNY SMITH APPLES.

Storage tests with apples of this variety from the same orchard at Elgin were carried out in 1938, 1939 and 1940. The results for the first of these years have already been published (12). It was established that the variety shows two types of superficial scald ; one, the frigescence

* Apples in plain wrappers at 0° C. were mostly *337-336*.

type, was at its maximum at $-0 \cdot 6°$ C., the other appeared later at $2 \cdot 8°$ C., these being respectively the lowest and highest temperatures used. The frigescence type decreased in prevalence and intensity with increased temperature, the existence of the other type was merely indicated.

In 1939 the storage temperatures were $-1 \cdot 7°$ C., $0°$ C., $1 \cdot 7°$ C. and $3 \cdot 3°$ C. The results plotted from the end of the third month onwards only, are illustrated in Fig. 3, A. After two months the number of affected apples did not exceed 4 per cent., and there was then no clear relationship between their number and the temperature. The curves show a decrease in frigescence and an increase in superficial scald with increase of temperature, the former developing most actively earlier than the latter.

The evaluation of the superficial scald was more difficult than that of the frigescence type. At the end of four months storage and thereafter, the latter was evident as dark brown, slightly sunken areas. In the former there was a gradual merging from a dull appearance to a dirty green and grey discoloration, which, in turn, merged into a dirty light greyish brown colour. Apples showing unmistakable discoloured areas were regarded as scalded, a proceeding which

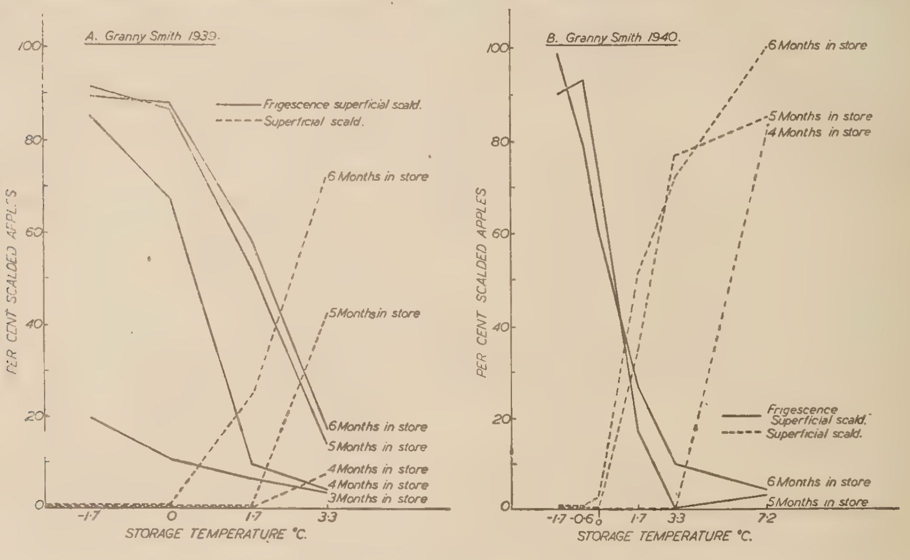

FIG. 3.

The superficial scalds (prevalence) of Granny Smith apples in relation to the temperature and length of storage.

would seem to be an extension of the term superficial scald as generally employed, although a limited number of the apples showing greyish brown discoloured areas might be described as showing superficial scald. This matter will be dealt with later, in the discussion.

The existence of two types of superficial scald leads inevitably to difficulties in estimating the exact prevalence of each, but this does not apply to the frigescence type at its range of optimum development. At the higher temperatures, however, it may sometimes be difficult to decide whether an apple shows slight scald of the frigescence type or severe superficial scald, and this difficulty tends to increase with increase of temperature. The estimation of the less severely discoloured apples also presents difficulties, but the general trends and relationships are clear. The irregularities recorded are due in part to such difficulties but they are not very serious, particularly with frigescence superficial scald.

Comparison of apples in plain and oiled wrappers after four months showed that there was less frigescence superficial scald with the latter than the former, and this was true after six months also, except at $-1 \cdot 7°$ C. The detailed figures are given in Table IV.

TABLE IV.

The effect of oiled wrappers on the prevalence of frigescence superficial scald and superficial scald in Granny Smith apples (1939).

Months in store (+1 week at 18·3° C.)	Storage temp. ° C.	Per cent. frigescence superficial scald.				Per cent. superficial scald.	
		Plain wrappers.	Oiled wrappers.	Double oiled wrappers.	Double oiled wrappers left intact at 18·3° C.	Plain wrappers.	Oiled wrappers.
4	−1·7	85·0	45·5	—	—	0·0	0·0
	0·0	67·0	12·6	—	—	0·0	0·0
	1·7	9·3	8·1	—	—	0·0	0·0
	3·3	3·3	1·8	—	—	5·7	0·0
6	−1·7	89·5	88·5	94·6	97·6	0·0	0·0
	0·0	87·5	15·9	24·8	13·6	0·0	3·2
	1·7	58·0	8·3	—	—	24·4	27·5
	3·3	17·2	0·0	—	—	72·0	74·1

The use of two wrappers and the retention of both at 18·3° C. was not beneficial, but rather the reverse, although the number of tests was not large enough to settle the point conclusively. Oiled wrappers, on the whole, were more effective in 1939, and although they reduced the intensity of superficial scald they did not reduce its prevalence.

The results obtained in 1940, when storage at 7·2° C. was also employed, are plotted in Fig. 3, B. Both types of scald developed, one at the lower temperatures, −1·7 C. and 0° C., and the other, later, at the higher ones, 3·3° C. and 7·2° C. Oiled wrappers controlled the frigescence type better than in 1939 and still better than in 1938; they had reduced the prevalence of superficial scald at 7·2° C. at the end of four months, but had no effect on it at the end of five; its intensity was reduced for both periods. The figures are given in Tables V and VI.

At 7·2° C. superficial scald was evident at the end of four months storage, its prevailing colour being a dirty greenish brown (*223*). The prevalence of superficial scald after five months had not appreciably altered though its intensity had increased, and its prevailing colour was a dirty light greyish brown (*262*). At this time many of the unscalded apples were of a dull colour, and at the end of six months all the apples showed superficial scald.

TABLE V.

Effect of oiled wrappers on the prevalence of frigescence superficial scald and superficial scald in Granny Smith apples (1940)

Months in store (+1 week at 20° C.)	Storage temp. ° C.	Per cent. frigescence superficial scald.		Per cent. superficial scald.	
		Plain wrappers.	Oiled wrappers.	Plain wrappers.	Oiled wrappers.
4	−1·7	28·0	14·7	0·0	0·0
	0·0	26·0	0·0	0·0	0·0
	7·2	2·1	0·0	82·0	56·8
5	−0·6	86·0	12·6	0·0	0·0
	7·2	2·9	0·8	85·0	86·0
6	−0·6	79·0	12·5	0·0	5·9

Table VI.

The development of superficial scald Granny Smith apples stored at 3·3° and 7·2° C.

Months in store. (+1 week at 20° C.)	Per cent. superficial scald.	
	At 3·3° C.	At 7·2° C.
4	0·0	82·0
5	76·5 (plus 11 per cent. of a dull colour)	85·0
6	72·0 (plus 19·1 per cent. of a dull colour)	100·0

After four months storage at 3·3° C. many of the apples appeared dull, but definite discoloration occurred only after five months, when grey-green hues prevailed. At the end of six months, fewer apples were affected at this temperature than at 7·2° C. and dirty, greenish-grey and grey colours still predominated. The effect was more in the nature of a discoloration at 3·3° C. as compared with the superficial scald at 7·2° C. This was particularly so at the end of five months, and thus the values at 3·3° C. given in Table VI do not fully represent the difference in the condition at the end of the fifth and sixth months of storage.

Superficial Scald in Rome Beauty Apples.

Tests were made with apples from Vereeniging in 1937, from Vereeniging and the Langkloof in 1938, from the Langkloof in 1939 and from the Langkloof and Elgin in 1940. The results for 1937 and 1938 have already been published (10, 11), those for 1939 and 1940 present a much more complex picture. More data are required for this variety than have yet been obtained, but discussion of the results for other varieties necessitates a consideration of the general features of those for Rome Beauty.

During 1937 and 1938 only small, circular brown areas surrounding lenticels ("spotting" type) and larger or smaller areas of a medium brown colour, usually close together, were distinguished, the spotting being more characteristic of the fruit stored at 2·8° C. and 3·3° C. In 1937 the relation between the number of scalded apples and temperature was not clear, owing to the high prevalence of superficial scald at 0·6° C., 1·7° C. and 3·3° C. On the whole it was greater at the higher temperatures. Serious development of it was evident only after four months in store ; thereafter, its prevalence and intensity increased with length of storage. On the whole the non-spotting type increased with decreased temperature.

In 1938 the prevalence of superficial scald was still high, but it was less severe and its temperature relationships were again not clear. It was most severe in the Vereeniging apples at 1·1° C. In those from the Langkloof it was more prevalent during the first four months at −0·6° C. and 1·1° C. than at 2·8° C., the point at which prevalence was greatest after five months. As in 1937, the non-spotted scald was better developed at the lower temperatures (0·6° C. and 1·1° C.) than at 2·8° C. Oiled wrappers eliminated superficial scald almost completely.

In 1939 the prevalence of superficial scald was of the same order as in 1937, it increased with increase of temperature from −1·7° C. to 3·3° C. Oiled wrappers completely eliminated or greatly reduced it, except at 3·3° C., at which temperature they exerted only a small effect after four, and none after five months storage.

Distinguishing between non-spotted and spotted superficial scald, the following statements hold good :

(1) After three and four months storage the spotted type was absent at −1·7° C. and present only to a limited extent at 0° C. Its prevalence increased consistently with increased temperature.

(2) After five months at 3·3° C. it was replaced almost entirely by dirty green and the usual brown discoloured areas, which, in many instances, had clearly arisen by the merging of circular ones.

(3) The non-spotted type was at a maximum at 1·7° C. after three months, but a month later it was of the same magnitude at 0° C. as at that temperature. There was a definite decrease both with increased and decreased temperature, more marked with the latter.

(4) Oiled wrappers (except at 3·3° C.) either completely eliminated superficial scald or reduced its prevalence very considerably, the non-spotted form more completely than the other. At 3·3° C. the number of scalded apples remained about the same whether plain or oiled wrappers were used. The non-spotted form, however, was entirely replaced by the spotted one with apples in oiled wrappers.

In 1940 apples from the Langkloof were stored at 0° C., 1·7° C., 3·3° C. and 7·2° C., whilst others from Elgin were stored at these temperatures and also at −1·7° C. From three months onwards the prevalence of superficial scald was high in both lots at all temperatures except −1·7° C. The fruits from Elgin showed consistently more of it at 7·2° C. than at 0° C. and −1·7° C., and those from the Langkloof more at both 3·3° C. and 7·2° C. than at 0° C. Maximum development was at 1·7° C. or 0° C., usually (and always with the Langkloof lot) at the former temperature.*

Oiled wrappers markedly lessened the scald after four months storage, except at 1·7° C., when they were less effective than in previous seasons and at which point its prevalence was the same for both plain and oiled wrappers. This, and the occurrence of the maximum prevalence and intensity at this temperature, indicated it as being the one of the optimum development of superficial scald in the Langkloof apples tested. On the whole, the Elgin apples showed increasing intensity of this scald with increasing storage temperature.

Distinguishing again between spotted and ordinary superficial scald, the tendency is once more seen for the maximum development of the latter to occur at 1·7° C., with a very definite decrease at 7·2° C. The prevalence of the former increased with increased temperature. It may be added that in very severe superficial scald the flesh is sometimes affected for a depth of a few millimetres below the skin.

Superficial scald in Rome Beauty was more complex than indicated above, since the colour of the ordinary form varied and included dirty green, light brown and medium brown. Many of the apples, too, showed a combination of two or more of these variations, and analysis of the temperature relations of the chief variations was rendered difficult and uncertain.

The relative increase in the spotted form and in the green, non-spotted, superficial scald effected by oiled wrappers, and their failure at 1·7° C. to control the development of the darker, brown coloured variation, suggest that the differences in appearance may be quantitative; and this is supported by the greater proportion of apples showing spotted superficial scald immediately on withdrawal from storage, before transference to 20° C.

The Superficial Scalds of Wemmershoek and White Winter Pearmain Apples.

The results of storage tests with Wemmershoek apples from Villiersdorp in 1937 have already been published (11). Superficial scald developed chiefly at −0·6° C., negligibly at 2·8° C., though the apples were very dull in appearance. It was of the frigescence type. With the same variety from Villiersdorp and Zuurbraak in 1939 similar results were obtained, though the prevalence was lower. Oiled wrappers completely eliminated or considerably reduced this frigescence superficial scald.

* At the end of four months the value for total superficial scald in the Langkloof apples was the same at 1·7° C. and 7·2° C.

The storage behaviour of White Winter Pearmain apples has been studied during four consecutive seasons, but more particularly in 1939 and 1940. The scald developed late in storage life, was practically absent at $-1 \cdot 7°$ C. and $0°$ C., but occurred at higher temperatures. Fruit from Elgin, showed little or no scald, except at $7 \cdot 2°$ C., at which point about 22 per cent. of it was recorded in 1940. Scald occurred to a greater extent in apples from the Langkloof in 1939 and 1940. In one instance 56 per cent. was recorded but otherwise it did not exceed 40 per cent. and its intensity was mostly low. It was of the spotted variety with a tendency to increasing confluence of the spotted areas.

Discussion.

Superficial scald of the first type (as defined here), characteristic of higher storage temperatures, most prevalent and typical late in the storage period and exhibited by the Ohenimuri apples, seems to correspond most nearly to the scald (2, 5, 13, 18, 19, 22), apple scald and superficial scald mentioned in the literature. The type here called frigescence superficial scald, occurring mainly at lower temperatures ($-1 \cdot 7°$ C. to $1 \cdot 1°$ C.), though not necessarily increasing in prevalence and intensity with fall of temperature over this range (cf. Red Delicious), has been reported occasionally during recent years (8, 9, 11, 12).

It is difficult to include in either of the above types the scald developed by Rome Beauty, since the temperature at which its optimum development occurred varied, and it was well developed at all the temperatures employed except $-1 \cdot 7°$ C. Possibly two types existed here, a medium brown non-spotted form, mainly at lower temperatures, and the spotted one at higher ones ; the latter is definitely a high temperature superficial scald. The most likely explanation is that superficial scald is more affected by pre-storage treatment in this variety than in the others here studied. For practical purposes Rome Beauty scald may perhaps be regarded as a third type, one chiefly determined by pre-storage factors.

As to the nomenclature of the two types, superficial scald, since it appears late in storage, might perhaps be termed senescence scald. In the Ohenimuri apples it recalls the " senescence characteristics " described by Thomas and Fidler (21) for Newtown Wonder and Bramley's Seedling apples, though, on the whole, the senescence features are not always well marked. Brooks, Cooley and Fisher (3) concluded that superficial scald was not purely an old age characteristic. Hence, although the cumbersome term " storage senescence superficial scald " might be applicable, it seems best, for the present, to retain the name superficial scald for the first type described here. The term frigescence superficial scald is proposed for the second type, which occurs at low temperatures, retention of the word superficial being necessary to distinguish it from the deep or soft scald considered by Plagge and Maney (17) to be a form of soggy breakdown.

Perhaps the chief difference between superficial scald as defined above and the scald referred to in the literature as developing at higher temperatures, is that skin discolorations are taken into account here, whereas they may not have been included by other workers or may have been described as late scald (6). This applies more particularly to the superficial scald of Granny Smith and, to a less extent, to Ohenimuri apples at $3 \cdot 3°$ C. Still, dullness (Wemmershoek) grades into discoloration (Granny Smith) and the latter to light brown superficial scald (Ohenimuri), and these seem essentially to correspond, though the dullness of Wemmershoek is so indefinite that it can hardly be included under the term superficial scald.

It should be noted that normal apples grade into dull ones, then to discoloured and finally to scalded ones, e.g. in Ohenimuri and sometimes Rome Beauty. Dullness or discoloration appearing early in storage may become superficial scald later. The use of oiled wrappers sometimes changes a superficial scald into a discoloration. Such facts suggest that the difference between superficial scald, in the sense of a browning of the skin, and skin discoloration (greyish green, dirty green, greyish-brown or very light brown colours) is quantitative, not

qualitative. The typical spotted superficial scald of Winter Pearmain and Rome Beauty cannot be described as an indefinite discoloration.

It is realized that the immediate chemical changes associated with the two forms of superficial scald described above may be the same for both. The current view is that scald is associated with the production of esters and acetaldyhyde ; if so it will be influenced by the quantity of such volatile products arising, their rate of production, the sensitiveness of the apple skin and the rate of elimination from the fruit. Nevertheless, the mechanism of production of the volatile substances may not be the same for both types of scald. In their work on zymasis and oxygen concentration Thomas and Fidler show that zymasis, and consequently alcohol production, is progressively retarded with increasing oxygen concentration. In old apples, however, alcohol may accumulate even in an atmosphere of pure oxygen. The failure of a 2·5 per cent. oxygen concentration to inhibit zymasis completely, coincides with the appearance of senescent characteristics, amongst which these authors include superficial scald. This may hold good for superficial scald as defined in the present paper, but frigescence superficial scald is a low temperature and not a senescence phenomenon.

Spotted superficial scald and Jonathan spot. The spotted superficial scald described above for South African apples agrees well with the description of Jonathan spot given by McAlpine (15), and in some respects with that given by Tiller (22) and Plagge *et al* (18) ; but the occurrence of Jonathan spot early during storage and sometimes even while the fruit is still on the tree distinguishes it from the spotted scald of South African apples. Other workers on Jonathan spot (4, 7, 23) have emphasized the necrosis of tissue beneath the expanding spots. Storage tests with South African Jonathan apples for one season have shown that they may be affected with spotted superficial scald like that of Rome Beauty and Winter Pearmain described above, but only to a very limited extent except at 7·2° C. Dark depressed areas overlying necrosed tissue, often appearing early during storage, were observed much less frequently in Jonathan and other varieties. So far as South African apples are concerned, spotted superficial scald is merely a form of superficial scald.

Frigescence superficial scald and soft scald. Soft scald, a recognized low temperature injury, may possibly be a very severe but more localized form of frigescence superficial scald. Soft scald has been produced typically in many varieties of apples by the same means as superficial scald. Which kind of scald will arise seems to depend on the variety, the toxic vapour and the length of exposure to it. Typical fully developed frigescence superficial scald in Granny Smith has not been induced artificially, but all the other variants of both types of scald have been so produced. In experiments carried out by Adam (1) a number of apple varieties were exposed to ethyl malonate vapour. The resulting injury was called apple scald, but two, if not three, of the apples illustrated show typical soft scald.

The temperature relationships of soft scald and frigescence superficial scald are similar, while in Rome Beauty the flesh was sometimes affected to a depth of a few millimetres. This is said to occur in severely scalded American apples (13, 19). Tests with oiled wrappers, taken as a whole, show that they had little or no effect on the incidence of soft scald ; and whilst they are generally useful in eliminating or reducing frigescence superficial scald their effectiveness varies and, as has been suggested above, is quantitative rather than qualitative. The severer the frigescence scald the less effective are the oiled wrappers.

Frigescence superficial scald and internal browning of Yellow Newtown apples. The internal browning of apples of this variety described by Overholser, Winkler and Jacob (16) may be related physiologically to superficial scald. According to these authors the browning is due to the accumulation of essential oils or other deleterious substances produced by the apples, and scald is closely related to it as to cause. The severity of the browning increased with decreasing storage temperature, from 7·2° C. to −1·1° C., while it was not serious at 7·2° C. or at higher temperatures, even after five months' storage. This suggests a closer affinity of internal browning with frigescence superficial scald than with superficial scald.

Summary.

Cold storage tests were carried out with six varieties of South African apples, at different temperatures, and the kind and amount of superficial scald present on them at the end of one week, after removal from storage, during which they were kept in a dark room at 18·3° C. or 20° C. with continuous air circulation, was determined. Two types of scald are distinguished :

(a) *Superficial scald*, tending to develop late in storage at relatively high temperatures (3·3° C. and 7·2° C.) and seen in the varieties Ohenimuri, Granny Smith and White Winter Pearmain. It approximates most closely to the scald, apple scald and superficial scald of earlier literature, but the term as here applied includes discolorations, not always included by other workers.

(b) *Frigescence superficial scald*, developed chiefly at lower temperatures (—1·7° C. to 0° C.), and well seen in the varieties Red Delicious, Granny Smith and Wemmershoek. The two last-named showed increased prevalence and intensity of it with decrease in temperature, the first one showed a maximum of both at 0° C.

In the variety, Rome Beauty, the character of the scald varied considerably, and did not readily fall within the scope of either of the above two types. It was very prevalent at all storage temperatures, that of optimum development varying with the season. On the whole it was more severe at higher than at low temperatures, and the spotted form of it was certainly more characteristic at the higher ones. It is suggested that in this variety the scald is affected by pre-storage conditions to a greater extent than in the other varieties studied.

The relationship of spotted superficial scald to Jonathan spot is discussed, as is also that of frigescence superficial scald to soft scald (which may be a form of it) and to the internal browning of Yellow Newtown apples.

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STUDIES IN THE NON-SETTING OF PEARS

VII.* THE GROWTH CYCLE AND FRUIT BUD DIFFERENTIATION OF CONFERENCE AND DOYENNÉ DU COMICE

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INTRODUCTION.

The small yield of fruit from some of the best varieties of pears has for many years been a bar to their commercial production. In the hope of finding a way to improve their performance research was begun at East Malling in 1931. It was continued there in greater detail by Srivastava (1938, a, b, c) who concluded that the non-setting of fruit in pears was a nutritional problem. The present paper deals with another aspect of the same problem, namely the growth cycle and fruit bud differentiation in the free-cropping variety Conference and the shy-cropping one, Doyenné du Comice.

The growth cycle was investigated because Lees (1926) suggested that the annual crop of apples was influenced by conditions during the preceding year. He showed that the size of the apple crop in any given year was very largely controlled by the size of the crop and the rainfall of the previous year, and suggested that these two factors determine the size of the future crop by their effect on the internal condition of the tree. A table was drawn up of the possible combinations of the two factors, with a forecast of their probable effect on the crop in the succeeding year. The accuracy of this forecast was checked from the records of the Woburn Fruit Farm and the Long Ashton Research Station, and except for certain years with spring frosts or serious aphis damage, the correlation between forecast and yield was good.

Many individual factors come into play for each tree, such as stock influence, manuring and pruning, but when large numbers of trees are used, these can be eliminated as being approximately constant from year to year.

The two factors mentioned were thought to affect the quantity of food stored in the tree during the vegetative period. It was considered that the organic and inorganic nutrients might be used by the tree for growth or be stored up as reserves. It is known that trees bearing a heavy crop makes less growth than those with little fruit, and also that trees make more growth during a wet summer than during a dry one. Since crop and rainfall affect the size of the succeeding crop, it would seem that growth requirements take priority over storage.

A similar conclusion as to the effect of growth on food storage has been deduced from direct measurements of the osmotic pressure of the cell sap. Measurements taken during dormancy, the growing season and before leaf fall, showed that the concentration was lowest during the period of most rapid growth (Chandler, 1925, p. 38). The problem is not a simple one, however, because as growth proceeds leaf area increases, consequently the increased products of photosynthesis may more than balance the amount of food required for growth. But it is clear that a tree which has ceased growth has a larger proportion of its food material available for storage than one which is still growing.

For these reasons, it seemed possible that a study of the growth cycles of a free and a shy-cropping pear might throw some light on one of the conditions which govern food storage, and hence of one of the factors which affect the set of fruit. For comparison, Conference, one of the most fruitful pears grown on a commercial scale, and Doyenné du Comice, a very shy-cropper, but one whose fruit has "that perfect combination of flavour, aroma and texture of which man

* For Part I, see *J. Pomol.*, 1938; Parts II and III, *E. Malling Ann. Rep. for* 1937; Parts IV, V, VI, *E. Malling Ann. Rep. for* 1940.

had long dreamed " (Bunyard, 1929) were chosen. Both are diploids (Moffett, 1934), so that differences in behaviour cannot be explained by differing chromosome numbers.

The study was mainly concerned with (1) the elongation of the leader shoots, (2) the number and time of development of the spur leaves and (3) the date of differentiation of the flower buds. In addition, in 1939 and 1940, spurs were labelled according to type, and their behaviour was noted in the succeeding year to determine whether the type of the parent spur influenced the type of the descendant bud.

Leader Shoot Growth.

Material.

Eight trees of Conference and eight of Comice were selected, four of each variety on Malling Quince A and four on Malling Quince C (Hatton, 1920). It was hoped that the results would indicate both varietal influence and rootstock effect.

Care was taken to choose Comice trees still on the quince roots, but the wide variation in vigour of those on Quince C suggested that scion-rooting might have taken place. The unions were therefore uncovered and two of these trees were found to have a single large scion root, but the other six were still on quince roots. When the data were analysed statistically the mean of the two remaining trees of Comice on Quince C was substituted for the missing ones, and for this reason deductions as to the behaviour of this combination are not so reliable as for the others. The Conference trees were not examined, since scion-rooting of this variety has not, as yet, been observed.

The Conference trees were eighteen years old in 1939 and the Comice one year younger, the trees having been planted as maidens. They were mature bush trees, trained with open centres ; leader pruning is now hard, generally back to from four to six buds. Young lateral shoots are spur-pruned back to three buds and compound spurs are thinned out when necessary.

The soil is a well-drained loam and belongs to the Malling series (Bane and Jones, 1934). It is clean cultivated during the growing season until the end of June or July. After that, weeds are left to grow until cultivation begins again in the spring, when they are either disced or ploughed in.

The following quantities of manure per acre have been applied during the last five years: from 1936 to 1940 inclusive, 2 cwt. of sulphate of potash per annum, and in 1938 and 1939 3 cwt. of superphosphate. The plot has received humus in several forms ; in 1937 12 tons of dung; in 1938 a crop of tares, ploughed in, together with 15 tons of humus from a compost heap ; in 1940 10 cwt. of high grade shoddy.

The trees have been routine sprayed with a tar-oil winter wash followed by lime-sulphur pre-blossom and Bordeaux mixture post-blossom sprays.

Procedure.

Five main branches on the south side and the same number on the north side were selected on each tree. All the new shoots from the one year old leader of the branch were measured weekly throughout the growing season. The term leader is used to describe the dominant shoot at the top of the branch. In the two varieties used the leader generally grows from the apical bud of the one year old shoot, but when this has been damaged the nearest bud sends out a shoot which becomes dominant. When this occurred the measurements of the dominant shoot have been used in calculating the amount of growth made. All side shoots on the one year old wood below the leader may be classified as laterals. The measurements, taken to the end of the shoot and not to the tip of the bunched up leaves covering the growing part, were made to the lowest whole centimetre, except those of small shoots between 0·5 and 0·99 cm. long, which were recorded as 0·5 cm.

To determine the time of cessation of growth the date when the uppermost leaf of each shoot

became unfolded was recorded. This usually coincided with the cessation of increase in shoot length, but in a few cases was a week later.

Three stages have been distinguished in the annual growth cycle of the pear (Barker and Lees, 1919):

1st Stage. After dormancy the buds start into growth, sending out a shoot of bunched leaves. This stage lasts whilst all the buds are making equal growth. In 1939, growth began about March 16th, and the end of this stage was reached on April 18th.

2nd Stage. The apical buds then grow more vigorously than those nearer the base, which form rosettes of leaves. This stage ends when these short spurs cease growth, which occurred on May 17th, in 1939.

3rd Stage. During this stage only the apical shoots continue to grow. In 1939 the Conference leader shoots finally ceased growth on July 25th, while those of Comice finished on August 28th.

The dates for the various stages at East Malling in 1939 agreed closely with those reported by Barker and Lees for the pear Vicar of Winkfield at Long Ashton in 1918, in spite of the differences in soil and climatic conditions at the two Research Stations. In 1940, when the growing period was much shorter, agreement with the Long Ashton dates was not so close.

The measurements of the shoots and spurs were started on May 3rd in 1939 and on May 1st, in 1940, both dates being approximately in the middle of Stage 2. The records were continued until the end of the third stage, in 1939 on August 28th and in 1940 on August 1st.

Results.

The mean length of the growing shoot was calculated from the weekly measurements and the figures were plotted to produce the curves in Fig. 1, p. 28. All the means were obtained from the measurements of forty shoots of each variety on each rootstock, with the exception of Conference on Quince C in 1939, with thirty-nine shoots, and Comice on Quince C, with twenty shoots for each year.

Statistical analysis showed that:

(*a*) There was no significant difference in the amount of growth made by the two varieties. In 1939, at the end of the growing season, the mean length of shoot of Comice on Quince A was 29·03 cm. and of Conference on Quince A 22·98 cm.; but in 1940 the order was reversed, Comice making a growth of 12·38 cm. and Conference of 16·39 cm.

(*b*) Although both varieties made greater growth on Quince A than on Quince C the difference was not quite significant at the 5 per cent. level. In the orchard, however, trees on Quince A grow to a larger size than those on Quince C, so that the difference in growth on the two rootstocks is more likely to have been due to rootstock influence than to chance.

With maiden trees the stock effect was reversed, for Conference trees on Quince C were found to make more growth than those on Quince A (Cannon, 1941). The shape of the growth curves for both the young and the mature trees was the typical S one, but with the difference that the young trees had a much longer growing period than the old ones.

(*c*) The difference in the mean shoot growth made in 1939 and in 1940 was most striking and was highly significant. In 1939 it was 21·3 cm. and in 1940, 11·7 cm. (Sig. Diff., $P=0 \cdot 05$, 4·2.)

The records of the date of cessation of growth were analysed statistically by assigning to each date its number of days from January 1st; thus July 25th, 1939, was the 206th day of the year. The analysis showed that:

(*a*) The difference in the dates on which Conference and Comice ceased growth was a significant one. The observations given in Table I under the heading Variety Means, show that Comice continued to grow nine days after Conference had ceased.

There was no obvious difference between either the varieties or the stocks when shoot growth began, so that the difference of nine days is probably a real one in the length of the growing season.

TABLE I.

The mean date of cessation of shoot growth in 1939 and 1940.

Variety.	Quince A.	Quince C.	Variety Means.	Sig. Diff. (P=0·05)
Conference	July 3rd (185·8)	July 3rd (186·0)	July 3rd (185·9)	8·4
Comice	July 22nd (204·6)	July 2nd (184·5)	July 12th (194·6)	
Stock means	July 12th (195·2)	July 2nd (185·3)		
Sig. Diff. (P=0·05) ..	8·4			

The significant difference between Comice on Quince A and Quince C exceeds 13·5. The figures in brackets represent the number of days from the first of January.

(*b*) Trees on Quince C ceased growth earlier than those on Quince A. This is shown by the Stock means in Table I.

(*c*) The rootstock effect on the date of cessation of growth is limited to Comice, for Conference stopped growing at approximately the same time on both rootstocks. This is shown in the Variety Means column of Table I, which also shows that Comice on Quince A continued growing about nineteen days longer than Conference.

It is clear that the rootstock effect on the length of the leader shoots of Conference did not arise from any difference in the date of cessation of growth, since shoots on both rootstocks ceased growth on July 3rd, but was due to an earlier slowing down in the rate of growth of trees on Quince C as compared with that of those on Quince A. This change in the rate of growth is clearly shown in Fig. 1.

(*d*) Growth ceased thirty-seven days earlier in 1940 than in 1939. The year means were: 1939, July 26th (208·9 days); 1940, June 20th (171·6 days); (Sig. Diff., P=0·05, 9·2).

SPUR GROWTH.

Material and Procedure.

The records were obtained from the same trees as those used for the measurement of leader growth and were made on individual spurs, usually part of a compound one carried on one of the main branches. The spurs were chosen at random along the length of branches on the north and south sides of the tree. They were divided into two categories according to Vyvyan's (1935) method for recording the summer growth of apples:

1. Primary growths from buds formed the previous year containing only embryonic leaves.
2. Secondary growth from a bud in the axil of a leaf of the current year.

Thus, leaf buds formed primary growths and flower buds secondary growths. The two types are shown in Fig. 2, Plate I, the spur on the left being a primary growth, that on the right a secondary one, the latter having grown from a bud in the axil of a leaf borne directly on the bourse.

Twenty primary and twenty secondary spurs were labelled on each tree, ten of each on the north and south sides. When the leaves were full grown the number of leaves on each spur was counted. Leaves on the bourse of secondary spurs were not included. From April 19th to August 23rd, 1939, and from May 1st to June 27th, 1940, a weekly record was also made of the condition of the spurs which were considered to have ceased growth when the last leaf uncurled.

In summarizing the results, any spur making a growth of more than 5 cm. was omitted. The distinction between a shoot and a spur is an arbitrary one, but in practice 5 cm. is a convenient limit, most of the spurs making a growth of less than 1 cm.

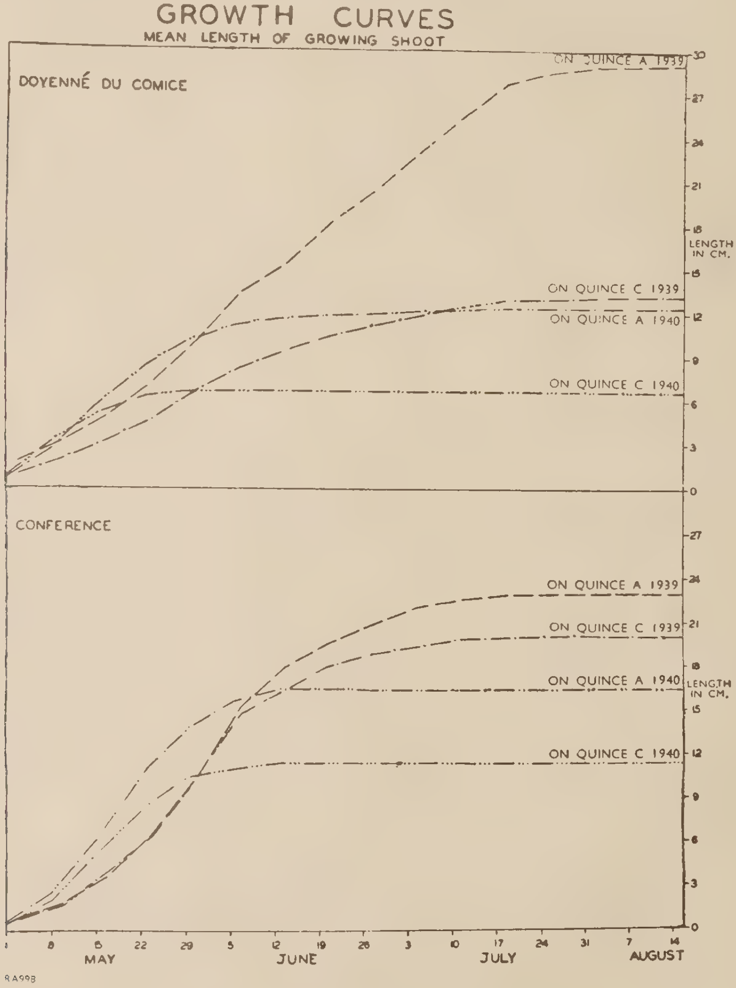

FIG. 1.

General Observations.

In 1938 the crop was small owing to a late spring frost, hence there was no check to fruit bud differentiation in that year, but in 1939 both varieties flowered abundantly, set well and gave an excellent crop. In 1940 there were fewer flowers, the difference between the years being much more marked with Comice than with Conference. The effect of the amount of blossom

on the leaf area of the tree was particularly striking with Comice, for in 1939 this variety appeared to be thinly clothed with leaves, whereas Conference was well covered. In 1940 Conference looked much the same as in the preceding year, but Comice had much improved, having plenty of large leaves. Other observers have also noticed " that the leaves are smaller and the total leaf surface considerably smaller in the year when the tree is bearing a heavy crop " (Chandler, 1925, p. 222).

Results.

In 1939 only a visual estimate was made of the proportion of primary to secondary spurs, from which it looked as though roughly 10 per cent. of the spurs of both varieties were primary. In 1940 an estimate was obtained from a sample count in which it was found that the proportion of primary growths had risen to 64·5 per cent. for Conference and 80·4 per cent. for Comice. These figures were calculated as the percentage mean from four trees of each variety on Quince A. The difference between the varieties is not significant at the 0·05 level, in all probability owing to the small number of replicates ; but the figures do show the great increase in the number of primary spurs in 1940 as compared with 1939.

Analysis of the number of leaves on the two types of spurs showed that :

(*a*) There was no significant difference in the number of leaves per spur due to varietal or stock effect. Primary and secondary spurs were selected in equal proportion, whereas as has just been shown, the proportion varied from year to year and possibly between varieties. This change in the proportion might produce differences in the total number of leaves on Conference and Comice.

(*b*) There were significantly fewer leaves per spur in 1940 than in 1939. The year means were : 1939, 3·83 ; 1940, 3·57 (Sig. Diff., P=0·05, 0·16).

(*c*) There was a suggestion that the difference between the years was most marked in Comice on Quince C, less for Conference on Quince A and not significant for Conference on Quince C and Comice on Quince A ; but triple interactions are difficult to interpret. The results are given in Table II.

TABLE II.

Effect of the triple interaction of variety, stock and year on the mean number of leaves per spur in 1939-40.

	1939.	1940.	Sig. Diff. (P=0·05)
Conference on A	3·93	3·55	0·31
Conference on C	3·81	3·94	—
Comice on A	3·75	3·60	—
Comice on C	3·81	3·19	0·43

(*d*) There was a significant difference in the number of leaves borne on a primary and secondary spur, the former having more leaves than the latter. This will be seen in section 1 of Table III.

TABLE III.

Mean number of leaves per primary and secondary spur in 1939-40.

	Primary spurs.	Secondary spurs.	Sig. Diff. (P=0·05)
1. Type of spur mean	4·28	3·11	0·21
2. 1939	3·83	3·83	—
1940	4·74	2·39	0·30
3. Conference	3·97	3·64	0·28
Comice	4·60	2·58	0·32
4. Conference 1939	3·28	4·46	0·40
Conference 1940	4·66	2·83	0·40
Comice 1939	4·38	3·19	0·46
Comice 1940	4·83	1·96	0·46

The significant difference applies only to values in the same line.

In the orchard, primary and secondary Conference leaves appeared to be similar in size, but the secondary leaves of Comice looked much smaller than the primary ones. Exact measurements of the respective areas would provide useful information on this point.

(*e*) The number of leaves on the primary and secondary spurs was equal in 1939, but in 1940 there were more primary than secondary leaves per spur. This is shown in Section 2 of Table III.

(*f*) The number of leaves on primary spurs of both varieties was greater than on secondary ones, and the difference was more marked with Comice than Conference. This will be seen in Section 3 of Table III.

(*g*) The difference in the number of leaves on the two types of spur was more marked in 1940 than in 1939, and the difference was greater with Comice than Conference. This is shown in Section 4 of Table III, which contains the results of the triple interaction of variety, year and type of spur.

In 1939 most of the spurs were secondary, while in 1940 roughly only a quarter of them were of this type, the importance of Section 4 of the Table in the interpretation of the behaviour of the trees thus becomes apparent. It shows that the predominant type of Conference spur possessed more leaves than the other, so that the trees appeared to have plenty of leaves in both years. Comice primary spurs, on the other hand, have always borne more leaves than the secondary ones. Thus, in the year when the secondary spurs were dominant, leaves did not appear to be plentiful, but when the situation was reversed they were abundant.

The records of the cessation of spur growth were treated in the same way as those of leader shoot growth, and the results were analysed. In 1940 it was observed that once a spur had ceased growth, it did not grow again ; but in 1939 a small proportion of spurs on both Conference and Comice made second growth which reached its peak about June 20th and ceased on August 12th. For this calculation no account was taken of second growth. While it probably resulted from more than one factor, it was most pronounced on trees which had been most severely attacked by pear aphides.

The analysis showed that :

(*a*) There was no varietal or rootstock effect on the final development of the leaves.

(*b*) Spur growth ceased three days earlier in 1940 than in 1939. (Table IV, Section 1.)

TABLE IV.

Mean date of cessation of spur growth in 1939-40.

	1939.	1940.	Sig. Diff. (P=0·05)
1. Year mean	May 12th (133·9)	May 10th (130·9)	1·41
2. Conference	May 13th (135·4)	May 9th (129·7)	1·86
Comice	May 10th (132·3)	May 11th (132·1)	—

In Tables IV and V the figures in brackets represent the number of days from the first of January.

(*c*) The difference in the ultimate date of leaf opening in the two years as shown by the variety and year interaction (Table IV, Section 2) was significant for Conference, but not for Comice. It will be seen that Comice was 3·1 days earlier than Conference in 1939 and 2·4 days later in 1940, the 1940 date thus being a gain of 5·5 days on Conference.

(*d*) The leaves on the secondary spurs unfolded four days later than those on the primary ones (Table V, Section 1.)

TABLE V.

Mean dates of cessation of primary and secondary spur growth in 1939-40.

	Primary spurs.	Secondary spurs.	Sig. Diff. (P=0·05)
1. Type of spur mean ..	May 8th (130·4)	May 12th (134·3)	0·87
2. Conference	May 9th (131·0)	May 12th (134·1)	1·13
Comice	May 8th (129·8)	May 12th (134·5)	1·33
3. 1939	May 10th (132·4)	May 13th (135·3)	1·23
1940	May 7th (128·4)	May 12th (133·3)	1·23

The significant difference applies only to values in the same line.

(*e*) Comice leaves on secondary spurs reached full development five days later than those on the primary ones, but with Conference the interval was three days (Table V, Section 2.)

(*f*) The difference between primary and secondary spurs was more marked in 1940 than in 1939. The interaction of type of spur with the year, shown in Table V, Section 3, shows that in 1939 the secondary spurs were later by 2·9 days and in 1940 by 4·9 days than the primary ones.

PARENTAGE OF SPUR BUDS.

Explanation and Procedure.

In order to find out how the leaf and flowering spurs would develop in the succeeding year, celluloid tags indelibly marked were tied to each spur. In the springs of 1940 and 1941 records were made as to whether the labelled buds were leaf or flower buds.

Results.

The percentage of primary buds which developed from primary and secondary growths of the previous year was calculated and the results analysed. It was found that there were significant differences in the behaviour of the two varieties in both 1940 and 1941.

Table VI.

Effect of variety on the mean percentage of primary buds arising from primary and secondary growths.

Year.	Conference.	Comice.	Significance level.
1940	34	82	$P < 0{\cdot}001$
1941	88	62	$P < 0{\cdot}001$

From Table VI it will be seen that in 1940, following a year when most of the spur growths were secondary ones, Conference continued to bear a large proportion of flower buds, whereas most of the Comice buds were leaf ones. In 1941 the relative position of the two varieties was reversed, for while the percentage of leaf buds on both varieties was high, there were more flower buds on Comice than on Conference.

The effect of rootstock on the production of leaf buds is presented in Table VII which shows that only in 1940 was the effect a significant one, for in that year trees on Quince A produced a higher proportion of leaf buds than those on Quince C.

Table VII.

Effect of rootstock on the mean percentage of primary buds arising from primary and secondary growths.

Year.	Quince A.	Quince C.	Significance level.
1940	62	53	$P < 0{\cdot}05$
1941	72	78	$P > 0{\cdot}2$

Table VIII shows the effect of the type of parent spur growth on the descendant buds.

Table VIII.

Effect of parentage on the mean percentage of primary buds arising from primary and secondary growths.

Year.	Primary.	Secondary.	Significance level.
1940	59	56	$P > 0{\cdot}2$
1941	63	87	$P < 0{\cdot}001$

It will be seen that in 1940 approximately the same percentage of primary buds arose from primary and secondary parent growths, but in 1941 the secondary growths produced a significantly larger percentage of primary buds than the primary parents. The matter can be summarized by saying that the type of parent spur influenced the character of the descendant buds in 1941 but not in 1940.

These results are of interest because it has been observed with apple spurs that blooming, if it has such a tendency at all, inhibits fruit bud formation by a spur only slightly, provided that no fruit is set (Chandler, 1925, p. 227). The percentages given in Table VIII are not strictly comparable with the apple observations because no distinction was made between fruiting and

non-fruiting spurs; but as the proportion of secondary spurs bearing fruit was small, the difference between the apple and pear observations probably cannot be entirely explained by this fact. The anomalous results of the effect of origin in 1940 and 1941 suggest that the subject would repay further study, particularly as it might throw more light on one of the factors affecting fruit bud formation.

THE DIFFERENTIATION OF THE FLOWER BUDS.

Explanation.

The time of differentiation of the flower buds has been determined for many kinds of fruit trees in all parts of the world, one of the objects being the discovery of the latest date at which any treatment designed to affect fruit bud formation will cease to be effective. Any treatment to alter the number of fruit buds must be applied before the physiological differentiation of the bud begins, because this precedes the morphological one.

In England, the date of differentiation of apple buds has been worked out by Gibbs and Swarbrick (1930), but there is, as yet, no published date for the pear in this country. With apples it has been shown that there are marked differences between varieties as to the date on which differentiation begins (Bradford, 1915). It was hoped that the present study would show what relationship the date of differentiation bore to the growth cycle, and whether there were varietal differences in the date on which Conference and Comice flower bud formation began.

Material.

In both 1939 and 1940 the buds were collected from trees similar in age and growing under the same conditions as those described in the section on shoot growth. In both years buds were collected from the primary and secondary spur growths, already described. When collecting, those buds that appeared to be potential fruit buds were picked, their leaves removed and the buds placed in a preservative solution. In the first year the buds were put into formalin acetic alcohol, made by adding 5 cc. of glacial acetic acid and 5 cc. of formalin to 90 cc. of 50 per cent. alcohol (Chamberlain, 1935). In the second year they were preserved in 70 per cent. alcohol, as recommended by Bell and Facey (1937) for dissection purposes.

In 1939 Conference and Comice buds from trees on Quince A were collected weekly from June 5th to August 2nd. As the supply of Comice buds was running short it was necessary to change over to trees on Quince B for the period August 11th to September 22nd, but fortunately it was found that Comice had differentiated before the change was made, so that the results were not upset by the change. Each week, from three trees of Conference and two of Comice, eight primary and eight secondary buds were collected from the north and south side of each tree, a total of thirty-two buds from each tree.

In 1940 it was possible to collect all the buds required from trees on Quince A weekly, from July 2nd to August 19th. As dissection does not require so many buds, only four primary and four secondary ones were collected from both the north and south side of a single tree of each variety.

Procedure.

Three methods were used for investigating the structure of the bud: (1) Dissection. (2) Division and examination of the cut face by incident light. (3) Serial sectioning for making permanent preparations.

In 1939 the approximate date of differentiation was determined by method (2) and sections were then prepared from the buds collected during the differentiation period. As there appeared to be some difference in the dates on which Conference and Comice differentiated, the dates were checked in 1940 by dissection. Experience proved that dissection was better than division for obtaining preliminary information. Dissection also enables the growing point to be seen in three dimensions, whereas with both the other methods it can be seen only in two.

Dissection.

The bud was first cut to a length of from 1·0 to 1·5 cm. and mounted axially on a triple pointed holder* to prevent its rotation. The shorter the bud the better, as it keeps steadier while the scales are being removed, but the wood at the base of the bud must not be so short that the needles approach the vegetative point. The holder was placed with the transfixed bud projecting over the edge of a glass block on which it could be revolved. The base of the scales and the lower scales themselves were then cut away, the holder being rotated a little after each cut. The cut was made towards the handle to avoid pulling the bud off the needles. The inner scales were easily removed afterwards with a dissecting needle, and as they became smaller it was helpful to look at the bud through a large hand-lens. When only a few small scales were left covering the vegetative point, the bud was ready either for final dissection with fine needles under the microscope or for cutting into small cubes, preparatory to dehydrating, embedding and sectioning.

When examining the dissected bud under the microscope it was refixed on the needles so that it was possible to look at the vegetative point. Horizontal illumination was also useful as this enabled differences in level to be seen more easily. The tissues of buds kept some time in a preservative turn brown, staining, therefore, is not necessary; but with comparatively fresh material iodine dissolved in 70 per cent. alcohol was useful. This dried quickly, so that the parts of the bud stood out clearly in relief, without being obscured by reflections from the surface of a liquid. Dissection and examination could be carried out after a little practice in about ten minutes. Investigators who have used the dissection method for examining buds are listed by Micklem (1938).

2. *Division.*

The bud was placed on a cork anvil and cut longitudinally as nearly as possible through the growing point, four or five buds at a time being thus treated. If fresh, the buds required staining with a water-soluble dye, but if preserved, the brown colour of the tissues rendered this unnecessary for the examination of general structure. It was difficult to ensure that the cut went through the small vegetative point, and this is the chief drawback to this method. The halved buds were then placed face up in a press chamber filled with glycerine. If the cut had passed through the centre of the bud, a comparison of the two halves enabled a fair idea of the shape of the vegetative point to be obtained.

3. *Serial sectioning.*

After the bud scales had been removed the material was cut into small cubes having sides 2·0 to 3·0 mm., in order to facilitate the penetration of the dehydrating agents and paraffin.

The material was dehydrated in stages following Bell and Facey's (1937) methods, using butyl alcohol under reduced pressure for 24 hours at each stage. It was embedded in paraffin (M.P. 52° C.) and longitudinal sections 10μ thick were cut on a rotary microtome. The parts of the ribbon containing sections of the vegetative point were selected, floated on water on a slide smeared with Mayer's fixative, warmed and allowed to dry for at least 24 hours. Staining was with acid fuchsin, 1 per cent. in 70 per cent. alcohol, for three minutes. A few seconds in 85 per cent. and 95 per cent. alcohol and two minutes in absolute sufficed for dehydration. Passage through stages of xylol preceded permanent mounting in balsam. For further details see Chamberlain (1935). It was found that sections after adhesion to the slide with Mayer's fixative could be kept in good condition unmounted for nearly a year.

Identification of Differentiated Buds.

Bell and McLellan (1939) enumerated various earlier criteria for the identification of the earliest stages of flower formation in apples, and described a more accurate one. They found

* Made by soldering the eye halves of a bundle of three medium sized sewing needles, whose pointed halves had previously been wound round with cotton thread to keep them apart, into a copper collar, afterwards provided with a wooden handle. Before use, the cotton thread was removed.

that the shape of the crown during the formation of bracts and leaf primordia was roughly triangular, but when flower formation began "the shape of the crown as seen in transverse section changed from triangular to circular". Pear flowers differ from apples in the order in which the parts of the flower are formed. In the apple the five sepal primordia of the king flower appear first, but in the pear a leaf or bract primordium at the base of the floral axis is the first morphological indication of flower formation. It was found, however, that the change in the shape of the crown in the pear provided an excellent dividing line between undifferentiated buds and those in which flower formation had just begun.

With serial sections the plan view of the crown cannot be seen unless models are constructed, so that other indications of differentiation must be used. Four points were found to be helpful in this connection :

(1) The maximum width of the crown as measured between the bases of the innermost bud scales is greater in differentiated than in undifferentiated buds. This was revealed by measurement of ten undifferentiated buds of Conference collected on June 28th, 1939, in which the maximum was 136 μ, the minimum 85 μ and the mean 97 μ, whilst the differentiated bud shown in Fig. 4, Plate II, collected on the same day, had a crown 220 μ wide. The difference is not always so large, however, for an undifferentiated Comice bud had a crown 187 μ wide while a differentiated one of the same variety measured 212 μ.

(2) The position of the widest point in the series of sections will sometimes indicate whether the crown is approximately triangular or circular in shape. For example, a Comice bud in which the sections had been cut parallel to the face of one of the innermost bud-scales gave the following series of successive crown-readings : 0, 68, 178, 170, 161, 161, 144, 127, 102, 76, 59, 17, 0 μ. The very rapid rise to the maximum width and the steady decline suggest that the sections had passed through a triangular crown, parallel to one side of the triangle. In another Comice bud the series was : 0, 187, 187, 204, 212, 195, 187, 178, 170, 170, 161, 161, 153, 144, 119, 102, 58, 42, 0 μ. Here the rise and descent at the ends are more gradual, the highest reading is nearer the centre, features consistent with a roughly circular crown. There is also a difference in the two series in the number of sections with widths over 100 μ, the undifferentiated bud having only eight as compared with fifteen in the differentiated one.

(3) The convexity of the crown above the base of the bud-scales is generally greater in the differentiated bud than in the undifferentiated one. The degree of curvature may be varietal, for it was more marked in Comice than in Conference in the early stages (Fig. 4, Plate II).

(4) The area of meristematic tissue is rather greater, and in longitudinal sections the bands of cells appear wider.

Results.

The results obtained by the three methods are given in Table IX, which shows that Conference flower buds begin to form towards the end of June, while those of Comice do not

TABLE IX.

Date of differentiation of buds in 1939-40.

Variety and stock.	Method of examination.	Bud examination began.	Differentiation first observed.
Conference on A	Bud division	5.6.39	5.7.39
Do.	Serial sectioning	28.6.39	28.6.39
Do.	Bud dissection	2.7.40	2.7.40
Comice on A	Bud division	28.6.39	2.8.39
Do. on A and B ..	Serial sectioning	12.7.39	11.8.39
Do. on A	Bud dissection	8.7.40	29.7.40

reach the same stage until the end of July. Although Comice buds begin to differentiate a month later than Conference, however, on the average the former variety comes into full bloom only four days later than the latter.

In 1939 only secondary buds were examined, because they were the predominant type. In 1940, when both primary and secondary buds were dissected, it was found that the former differentiated a little earlier than the latter, but the number of buds examined was not sufficiently large to determine whether there was a real difference in the date on which the two categories differentiate.

These results may be compared with those found in the northern hemisphere by other investigators who reported that pear buds of Wilder Early in Wisconsin differentiated on July 21st (Goff, 1899), of Kieffer in Virginia during mid-July (Drinkard, 1911), of Beurré Hardy in Holland on August 24th (Luyten, 1926), and of Williams' Bon Chrétien in California during the first week of July (Wiggans, 1925). A world list is given by Barnard and Read (1932).

A series of photomicrographs (Figs. 3-8, × 60, Plate II), was taken by direct projection on bromide paper to illustrate the early stages in the formation of the flower bud. The stages are described in the terms originally used by Barnard and Read (1932). Fig. 8 represents the most advanced stage found in all Conference buds up to August 2nd, 1939. No Comice buds were found in such an advanced condition, the most forward ones seen up to August 23rd, 1939, being still in stage II.

In each section some of the cells were seen to contain crystals, probably of calcium oxalate, which is frequently found in regions of active metabolism (Fritch and Salisbury, 1920). Although the buds had been collected at weekly intervals, no regular progression was shown by them. For example the bud in Fig. 7 was picked on July 12th, 1939, while that in Fig. 6, which represents an earlier stage, was picked nearly three weeks later, on August 2nd. It was not possible, therefore, to assign dates for the formation of the various parts of the flower, and the evidence suggests that with buds of secondary origin the differentiation of the flower bud may start over a period of several weeks.

The relation between the date of differentiation of the flower buds and the growth cycle is shown in Table X.

Table X.

Comparison of date of fruit bud differentiation with that of cessation of leader shoot and spur growth.

Variety (both on Quince A)	Year.	Date differentiation first observed.	Cessation of spur growth.	Cessation of shoot growth.	Mean date of cessation of shoot growth.
Conference	1939	June 28th	May 13th	July 20th	July 3rd
Conference	1940	July 2nd	May 9th	June 18th	
Comice	1939	Aug. 2nd	May 10th	Aug. 10th	July 22nd
Comice	1940	July 29th	May 11th	July 6th	

It will be seen that differentiation took place in Conference nearly eight weeks and in Comice approximately twelve weeks after spur growth had ceased. This is considerably longer than that reported for Williams' Bon Chrétien in Victoria (Australia), where the interval between the maturing of the spur leaves and the start of fruit bud formation was only two to three weeks (Barnard and Read, 1932). In South Africa, it was found that fruit bud differentiation began shortly after the cessation of growth of the lateral shoots on the main branches of the tree (Micklem, 1938). The results given in Table IX are not comparable with the South African observations because the measurements at East Malling were made on the leader shoots at the

top of the main branches of the tree. No definite relationship seems to exist, therefore, between the date of cessation of shoot growth and fruit bud differentiation in the two years 1939 and 1940; nevertheless the mean date for the two years is roughly the same as that for the beginning of fruit bud formation.

Discussion and Conclusions.

Comparison of the growth cycle of the free-cropping pear Conference with that of the shy-cropping Comice showed that there were important differences between the two varieties. It was found that the leader shoots of Comice on Quince A grew for nearly three weeks longer than those of Conference on both rootstocks. Growth curves also indicated that both the amount of growth and the length of the growing period were greater in 1939 than in 1940. Evidence obtained from other experiments on the same plot suggested that the small amount of growth in 1940 was not entirely due to lack of soil moisture. It therefore seems possible that the growth made in any given year is partly determined by the internal condition of the tree at the beginning of the vegetative season.

The observations on spur growth showed that the secondary spur leaves opened at a later date than primary ones. From this it follows that a high proportion of flowers, as shown by Comice during the " on " year, will delay the period of full leaf development and of maximum photosynthesis. They suggest that a tree with many flower buds must depend on its reserves for a longer period than one with a larger proportion of leaf buds. Moreover, the leaf area of Conference appeared to be approximately the same in both 1939 and 1940, while with Comice there was a marked change from small leaf area in the earlier year to large leaf area in the later one.

In both 1939 and 1940 flower formation was found to begin in Conference on Quince A towards the end of June, while Comice buds on the same rootstock started to differentiate about one month later. There is as yet no evidence to indicate the significance of this difference.

During the two years in which the comparison was made the free-cropping Conference had a shorter growing period and a larger leaf area in a year of high flower bud production than the shy-cropping Comice. The reduction of the leaf area of Comice probably led to a diminution of reserves, a condition which may well have been accentuated by its longer growing period.

SUMMARY

1. In 1939 and 1940 the growth cycle of mature Conference and Comice pear trees on Quinces A and C was studied by taking weekly measurements of the leader shoots and spurs throughout the growing season. There were four replicate trees of each variety on each rootstock. It was found that:

(i) Comice had a longer growing season than Conference.

(ii) Trees on Quince A made more growth than those on Quince C.

(iii) In 1939 there was more growth during a longer period than in 1940.

(iv) There were fewer leaves per spur in 1940 than in 1939.

(v) There were more leaves per primary spur (leaf bud growth) than on secondary ones (flower bud growth). Primary spur leaves also unfolded earlier than secondary ones.

2. By following the history of labelled spurs it was found that the percentage of primary and secondary spurs forming flower buds was approximately the same in 1939, but significantly different in 1940.

3. The date of differentiation of the flower-buds was determined in 1939 and 1940 by dissection, division and serial sectioning of presumed flower buds. It was found that the

Conference buds began to differentiate at the end of June, nearly eight weeks after spur growth had ceased, and those of Comice at the end of July, twelve weeks after the end of spur growth. A series of photomicrographs was taken illustrating the development of the flower up to the formation of the stamens, six of which are reproduced.

ACKNOWLEDGMENTS

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PLATE I.

R A 995

Fig. 2.

Primary spur growth on left, secondary on right.

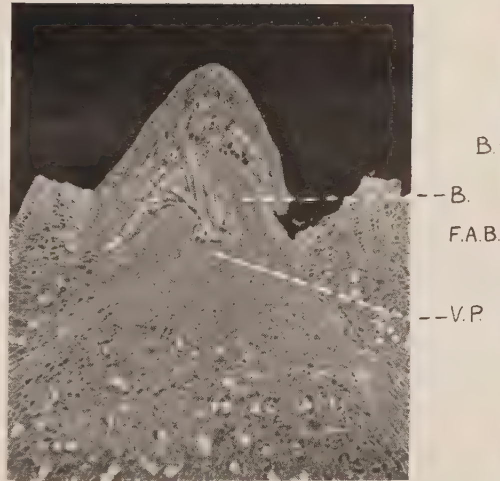

Fig. 3.

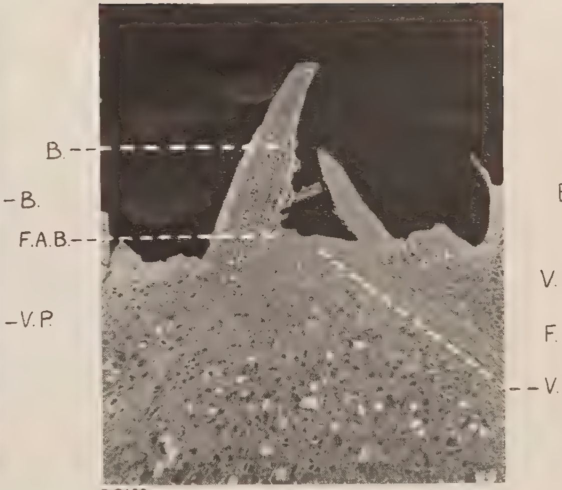

Fig. 4.

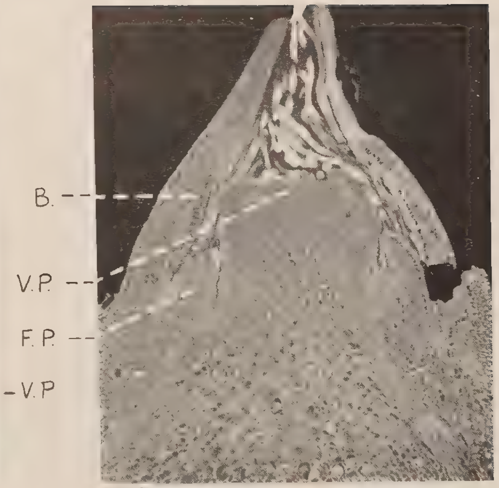

Fig. 5.

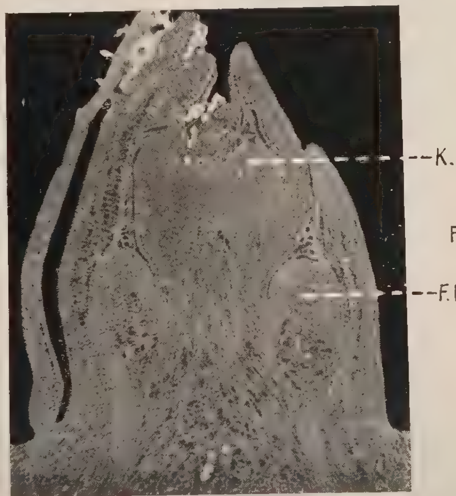

Fig. 6.

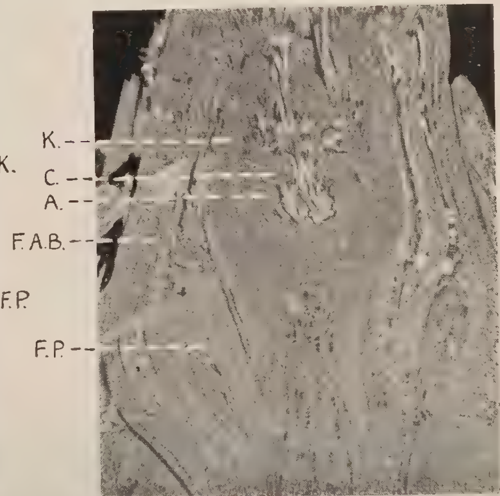

Fig. 7.

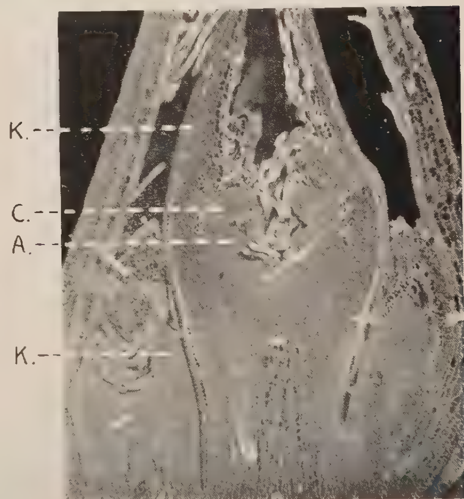

Fig. 8.

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EXPLANATION OF PLATE II

Development of the flower buds in the Conference pear.

Fig. 3.

Stage I. An undifferentiated bud with a small and nearly flat crown.

Fig. 4.

Stage II. The earliest stage of differentiation with a wide, slightly raised crown showing the first floral axis bract.

Fig. 5.

Stage III. The crown has flattened preparatory to the initiation of sepal growth in the King flower.

Fig. 6.

Stage IV. Sepal formation has taken place in the King flower.

Fig. 7.

Stage V. Both petals, and the first whorl of stamens are now present. The sepals are becoming hairy and continuing to grow.

Fig. 8.

Stage VI. Median section through the petals and second whorl of stamens.

Figs. 3, 4, 5 and 6, radial longitudinal sections; Figs. 7 and 8 tangential. All × 60.

Key: VP, Vegetative point; B, Bract; FAB, Floral axis bract; FP, Flower primordium; K, Sepals; C, Petals; A, Stamens.

(*Received* 1/9/41.)

PARTHENOCARPY INDUCED BY FROST IN PEARS

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Growers usually consider Conference and Fertility pears to be self-fertile, as judged by the fruit that sets in orchards consisting of these varieties only. Out of doors, under strictly isolated conditions, at the John Innes Horticultural Institution, Fertility produced seedless fruits following self-pollination, while Conference produced fruits which dropped off before fully maturing (Crane and Lewis, 1942). On the other hand, incompatibility tests in a cool greenhouse have shown that both these varieties are highly self-sterile. This discrepancy between outdoor and greenhouse results is not due to these varieties being self-fertile under some conditions and self-sterile under others, but to the fact that they are sometimes parthenocarpic.

It was noticed at the time of pollination of the outdoor trees that the styles of the flowers on many of them were blackened by frost, and it was thought that the action of the cold might have stimulated the development of parthenocarpic fruits. To test this idea, some of the branches of a potted tree of Conference were given cold treatments in a refrigerator, the others being left as controls. The treatments varied in duration, temperature, and stage of development of the flowers at the time of treatment. Furthermore, after treatment, some flowers were pollinated and others left unpollinated. The results are given in the following Table.

Table.

Results of exposing pear flowers to low temperatures.

Treatment.	No. of flowers.	Resulting damage.	No. of swollen fruits after 14 days.	No. of mature fruits.
0° C.—19 hrs.	41*	petals brown	25	0
−5° C.—18 hrs.	19	petals and styles brown	9	2
−7·5° C.—3 hrs.	48	Ditto.	21	0
−9° C.—17 hrs.	34*	18 fls. killed ; dropped off 16 damaged styles	9	0
Control	63	Nil.	0	0

* Except where marked with an asterisk the flowers were not pollinated ; half of these were pollinated after treatment and half not. No difference due to pollination could be observed.

Within a week after treatment a difference between the treated and the control branches became evident. The treated fruits had swollen while the untreated ones remained small and had begun to shrivel and fall. There was no apparent difference between the branches that had been subjected to cold treatment, whether pollinated or not. Many of the fruits on the treated branches continued to grow, but the majority of them fell off in June in a similar manner to those on a self-pollinated outdoor tree.

On the branch treated at −5° C. for 18 hours, unpollinated, two fruits reached maturity. These were not "thin" fruits but were good specimens, measuring 120 mm. long by 68 mm. in diameter. Neither fruit contained any seeds, and only traces of ovules were present.

The physiology of fruit development has been studied by Gustafson (1938), and parthenocarpic fruits have been produced in many plants by applying auxins to the ovary. The effect of cold may perhaps be to produce some auxin-like substance in the ovary which stimulates

fruit development. The only comparable effect of low temperature on the production of parthenocarpic fruits in other plants is that shown by seedless apples produced by frost (Whipple, 1912). Seedless apples of the variety Wealthy were produced by frost; but according to Latimer and Rawlings (1937), the embryos of the seedless fruits were killed by a frost that occurred three days after petal fall. This, therefore, was not a case of true parthenocarpy.

Cold-induced parthenocarpy may explain the discrepancies between results of different workers on self-sterility in certain varieties of pears [cf. Rawes (1933), Crane and Lewis (1942), Reinecke (1930)]; and the importance of considering the seed content as well as the percentage of fruit set, when estimating the self-sterility of a variety, is evident.

Parthenocarpy in pears is by no means uncommon, particularly after a spring in which frost has injured the flowers. In 1941 many fruits of seedling pears growing at the John Innes Horticultural Institution were examined, but only one fruit had any seeds. From observations of the seed content of a number of varieties, it appeared that some varieties responded to exposure to cold and others did not. Thus, cold stimulation may play a great part in the maintenance of a regular bearing habit in some pear varieties. In a cold spring, in which insects are not active to effect cross-pollination, a frost at the time of flowering, instead of doing damage, may be the making of a good crop.

SUMMARY.

Conference and Fertility pears are in some degree parthenocarpic and this condition is due to frost injury to the flowers. Parthenocarpic fruits were produced, without pollination, after treatment at $-5°$ C. for 18 hours at flowering time. This stimulation of parthenocarpy by cold may explain some of the discrepancies between the results of different workers on self-sterility in pears. It may also be of considerable importance in producing a crop of fruit in a cold spring as shown by the high proportion of seedless fruits formed in some years.

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APHIS TRANSMISSION OF STRAWBERRY CRINKLE IN GREAT BRITAIN

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Introduction.

Crinkle disease of strawberry was first described by Zeller and Vaughan (1932), who stated that it was prevalent in the North-Western States of America. Later, Vaughan (1933) and Zeller (1933) showed it to be a virus disease, and stated that it was transmitted by the strawberry aphis common in that area, *Myzus fragaefolii* Cel. It was first recorded in Britain in 1934 by Ogilvie, Swarbrick and Thompson (1934) who described the symptoms on Royal Sovereign plants from the South-west of England and from Worcestershire. The symptoms were stated to differ from those of Aphis plant (Briton-Jones and Staniland, 1927) and of Yellow-edge (Harris, 1933), but were considered to be identical with those of Crinkle described in North America. In 1935, Harris (1937), in a series of grafting experiments, showed that symptoms differing from those of Yellow-edge, together with those of Yellow edge itself, were transmitted by this means. He stated that the symptoms thus transmitted closely resembled those occurring naturally in the South-West of England and those of Crinkle in North-West America. By these and later grafting experiments Harris *et al* (Harris, 1937, 1938 ; Harris and Hildebrand, 1937 ; Harris and King, 1942) have shown that Crinkle is of widespread occurrence in this country and of considerable importance to the strawberry grower. Massee (1935) showed that the strawberry aphis (*Capitophorus fragariae* Theob.)* is a vector of Yellow-edge in this country. The frequent association of a mild form of Crinkle with Yellow-edge in Royal Sovereign, as shown by Harris and his co workers, suggested that this aphis might also act as a vector of Crinkle, and the experiments now to be described were made to test this possibility.

Aphis Transmission Experiments in 1937.

Healthy Plant Material. The experiments in the transmission of Yellow-edge (Massee, 1935) were made with plants from a clone of Royal Sovereign (Malling 35) known to be free from Yellow-edge but later shown to be affected with a mild form of Crinkle. It was therefore decided to use the common woodland strawberry (*Fragaria vesca* L.) which, in its natural habitat, was considered to be free from both Crinkle and Yellow-edge viruses (Harris and King, 1942). A single plant was selected from a local copse, transferred to a pot and kept in an insect-proof greenhouse. Runners raised from this one plant were used in the present investigation. Although the plants were grown in an insect-proof greenhouse, a Derris preparation was added to the water used for damping down the plants each morning and evening, as an additional precaution against possible inoculation by stray insects. Thirty-six plants of approximately equal size and vigour, available by the beginning of June, were divided into two equal batches of eighteen ; one of these was transferred to an adjoining insect-proof house.

Source of Inoculum and Aphis Material. A potted Royal Sovereign plant, exhibiting typical mild Crinkle symptoms, confirmed by a previous grafting test on healthy *F. vesca*, provided the source of inoculum. To this plant were transferred a number of adult apterous viviparous female aphides (*C. fragariae*) previously fed on a healthy *F. vesca* plant derived from the original clone. A strong colony of aphides soon became established on the diseased Royal Sovereign plant, and by the end of May the number required for inoculation purposes was available.

* It is considered by some authorities that *Capitophorus fragariae* Theob. and *Myzus fragaefolii* Cel. are the same species.

Aphis Transfers. On June 4th four apterous viviparous female adults were transferred by means of a camel hair brush to each of eighteen healthy *F. vesca* plants. Each aphis was placed on a partially opened leaflet at the centre of the crown of the plant. There were eighteen control plants and to each of these four adult apterous viviparous females from the colony reared on a healthy *F. vesca* plant were transferred. Subsequent observations showed that the aphides on both series of plants did not become established immediately, feeding not being observed for two days. During this period they wandered all over the plants but eventually started to feed on the petioles of the young leaves. They were left unmolested for one month, and by the end of that time strong colonies had become established on each plant. The aphides were then destroyed by completely submerging the plants in a solution of nicotine and soft soap. From this time onwards this dipping was repeated each week until the end of October, as a matter of routine. The plants were also dusted on three occasions with flowers of sulphur as a precaution against mildew, which is very prevalent when strawberries are grown in a greenhouse.

Observations on the plants. All the plants appeared to be quite healthy two weeks after the transfers had been made. Traces of leaf-edge curling were noted on the young leaves of plants of both series, but this is known to be caused by the mere feeding of the aphides and is not one of the initial stages of Crinkle symptoms. Similar leaf curling had been observed in previous experiments, and may be noted in any commercial strawberry field infested by aphides.

Traces of Crinkle symptoms were first noted on fourteen plants on June 20th, seventeen days following the direct transfers, and they consisted of translucent, chlorotic spots. At this date all the control plants were quite healthy, but the leaf curl already referred to was still visible. On June 26th Crinkle symptoms were more pronounced on these fourteen plants but the remaining four in this series were apparently still quite healthy. All the control plants were still normal. By the end of June all the plants in the series colonized with aphides from the diseased Royal Sovereign plant exhibited mild symptoms of Crinkle. The four which had previously appeared healthy now showed symptoms similar to the others. All the control plants were still quite healthy.

At the beginning of July the aphides on both batches of plants were destroyed by dipping in nicotine solution. Two runners from each plant of both series were allowed to root in pots adjacent to their parents, but were not severed from the latter, and from that time onward all other runners were removed from each plant as they appeared. On August 7th, all the plants in the series inoculated with aphides from the Crinkle affected Royal Sovereign plant still exhibited mild symptoms of Crinkle. At that time the intensity of symptom expression varied considerably amongst the plants, but in no case could it be described as severe. Each of the pairs of runners (still attached to the parent plants) exhibited mild Crinkle symptoms, and since none of them had been infested with aphides it is evident that the virus must have been transmitted to each along the stolon from the parent. All the control plants were still quite healthy at this time, retaining their healthy appearance and vigorous growth; the characteristic leaf curling recorded at the end of June was no longer visible, for the older leaves were hidden by the young growth. The runners from the control plants were also healthy and vigorous.

No further developments were observed during the autumn months and a final survey was made in the latter part of December, when the plants to which aphides from the affected Royal Sovereign plant had been transferred and their potted runners still attached to the parents continued to exhibit typical mild Crinkle symptoms. The older foliage was of a yellowish-green colour, while the young leaves were retarded and malformed. The control plants and their similarly potted runners, however, were still quite healthy, and the fresh green colour of their foliage contrasted notably with that of the plants in the Crinkle infected series.

The healthy *F. vesca* plant on which the original colony of aphides had been raised was still healthy at the end of December, although it had become considerably weakened by the continuous feeding of aphides on it throughout the year.

Conclusions. From the above results it is concluded that a virus inducing mild symptoms

of Crinkle was transmitted to healthy *F. vesca* plants by adult viviparous females of the strawberry aphis (*C. fragariae*) previously fed on a Royal Sovereign plant showing mild Crinkle symptoms. In this stage the strawberry aphis is therefore concluded to be a vector of mild Crinkle in this country, thus confirming the results of Zeller and Vaughan (1932) in America.

Aphis Transmission Experiments in 1938.

In 1938 the transfers were made a little earlier in the season with the immature stages of apterous viviparous females. The control plants in this experiment were not colonized with aphides. They were housed in a separate insect-proof greenhouse.

Source and Treatment of Healthy Plant Material. Twenty *F. vesca* plants raised from the original clone and growing in an insect-proof greenhouse were divided into two equal series, one being transferred to an adjoining insectary at the beginning of May. The blossom trusses and runners were removed from the plants of both series as they appeared. All the plants were dipped in a solution of nicotine and soft soap once each week until the time of the transfers, but from that time onwards the control plants alone were thus treated.

Source of Inoculum and Aphis Material. The colony of viruliferous aphides used in 1937 was again used, having survived the winter housed in a large muslin cage within a heated insect-proof greenhouse. The Crinkle-infected Royal Sovereign plant which had supported this colony of aphides continuously for well over a year was still sturdy, and typical mild Crinkle symptoms were still evident on it.

Transfers. On May 29th each plant was colonized with ten immature apterous viviparous females. The stages varied from the first to the fourth instars inclusive, but no attempt was made to colonize individual plants with specific instars. All the ten immature aphides were placed on a single leaflet of each plant, and they were observed to settle down and start feeding almost immediately. In the previous experiment the adult aphides then used did not start feeding for two days following transference. The aphides bred freely for a month and were then destroyed by the dipping in nicotine solution. The control plants were dipped once each week until the end of the experiment. Strong colonies of aphides had become established on each plant by the end of June, and they confined themselves mainly to the young foliage. The aphis population did not vary very much on the individual plants ; each supported about one hundred aphides, or more.

Observations on the Plants. On June 13th four of the colonized plants—three weeks after aphis transfer—showed incipient symptoms of mild Crinkle. They were detected on the young foliage only. By the end of June, some five weeks after transfer, the ten plants showed typical mild Crinkle symptoms. With few exceptions the leaf markings were confined to the young foliage, but scattered chlorotic spots were also present on the older leaves. The symptoms were more conspicuous on some plants, including the four which first showed incipient Crinkle, but the degree of infection was by no means severe.

On September 1st all the colonized plants exhibited more pronounced Crinkle symptoms than at the end of June, and the intensity of symptoms in each plant tended to equalize, rendering the plants much more uniform in appearance. Mild Crinkle symptoms were still apparent on each plant on November 9th, but in general the plants were in the same condition as they were in September. All the control plants remained entirely healthy throughout the experiment, and their upright habit and clear green foliage presented a striking contrast to that of the other series.

Height and spread Measurements. To obtain a measure of the difference in vigour of the plants of each series, height and spread measurements were made on May 25th, July 1st and November 9th. They are summarized in Table I.

It will be noted that it was not until November that any considerable difference between the vigour of the control and the colonized plants became evident. The apparent decrease

TABLE I.

Average Height and Spread Measurements in inches of F. vesca *plants in* 1938.

Treatment.	25.v.38. Height.	Spread.*	1.vii.38. Height.	Spread.*	9.xi.38. Height.	Spread.*
Controls	5	12	6·5	15·5	5·5	12
Inoculated plants ..	6	11·5	6	14·5	2·75	8

* One diameter only, at the widest part of each plant.

in the spread of both series between July and November was due to the usual autumnal dying off of the outer leaves.

Conclusions. The above results confirm those of the previous experiment and show that the immature forms of the apterous viviparous female *C. fragaria* are also vectors of the mild Crinkle virus. They also show that transmission can take place in May, the period in which this aphis spreads naturally from one strawberry field to another (Greenslade, 1941.).

APHIS TRANSMISSION EXPERIMENTS IN 1941.

A further experiment was planned to investigate the functions of the alate viviparous female stage as a vector of Crinkle virus, and the ability of this stage of the aphis to transmit mild and severe Crinkle in parallel trials.

Source and Treatment of Healthy Plant Material. In the previous experiments it was necessary to use *F. vesca*, owing to the lack of plants of the cultivated strawberry free from Crinkle. For the present experiment a clone of Royal Sovereign plants, proved by test to be free both from Crinkle and Yellow-edge, was available (Harris and King, 1942). Sixty-eight Royal Sovereign plants of this virus-free clone (Malling 40), raised in pots in an insect-proof greenhouse, were used. The blossom trusses and runners were removed as they developed. The plants were divided into three series, two of twenty-four plants each and one of twenty. Each series was kept in a separate compartment of the insectary. The twenty plants were used as controls.

Source of Inoculum and Aphis Material. The original strain of aphides used in the two previous experiments was used. A colony of them was established on a Royal Sovereign plant affected with mild Crinkle and another on a Royal Sovereign plant affected with severe Crinkle. These diseased plants were provided by Harris and King; they were identical in origin with Crinkle-affected material used in their study of this disease and were kept in separate cages in an insect-proof greenhouse. Strong colonies of aphides became established on each plant and by May 24th alate forms began to mature.

Transfers. The transfers were made on May 26th, 27th and 28th, since the alate forms were not present in sufficient numbers on any one day to supply all the plants at one time. A hurricane lamp glass, with fine muslin stretched over its top and fastened by a rubber band, was placed over each plant to prevent the aphides from escaping. This arrangement proved very efficient, since very few of the aphides were trapped in the drops of moisture which formed on the inside of the lamp glasses, and the daily routine of watering and caring for the plants was not impeded. The lamp glasses were removed as soon as all the winged forms had died. The alate forms were transferred immediately they became mature, but before their wings became fully expanded. Owing to the condensation of moisture on the inside of the lamp glasses it was impossible to see whether all the aphides settled down and fed on the foliage, but the majority of them were seen to become established and to thrive in spite of the confined conditions. They were allowed to breed on the plants for four months when, although they were still breeding,

the infestation was not large. They were destroyed on the plants by dipping and spraying, using a Derris preparation, in the last week of September.

The twenty control plants were not colonized with aphides ; they were kept in a separate compartment and dipped each week in a Derris preparation.

Observations on the Plants. Small colonies were establishing themselves on each plant by the end of June, showing that the winged forms must have settled down satisfactorily when they were transferred. The first appearance of Crinkle symptoms was noted on both series of colonized plants on July 1st. Three plants of those colonized with aphides from the mild Crinkle-affected plant, and two of those colonized from the severe Crinkle-infected plant, were then showing typical mild symptoms on the young foliage. During the second and third weeks of July several other plants of both series developed mild symptoms, and by the end of July every plant of both series was showing symptoms. By August 7th seventeen plants of the severe Crinkle series were exhibiting more severe symptoms than those visible in the plants of the mild Crinkle series. From the middle of August onward the symptoms in the severe Crinkle series became more pronounced, and by September 15th the contrast between the plants in the severe and the mild Crinkle series was extremely striking, both as regards symptom intensity and vigour of growth. All the plants in the series colonized with aphides fed on the mild Crinkle-affected plant were uniform in size and symptom intensity. Those in the corresponding severe Crinkle series (with the exception of one plant which died in August) were also uniform in some respects, but their vigour was very much less, and their symptom intensity greater than that shown in the plants in the mild Crinkle series (Plate I, Figs. 1 and 2). The control plants without exception retained their uniformly high vigour and freedom from Crinkle symptoms throughout the experiment (Plate II, Fig. 1).

Height and Spread Measurements. Height and spread measurements recorded on August 15th, September 15th, and October 8th are summarized in Table II.

TABLE II.

Average Height and Spread Measurements in inches of Royal Sovereign Strawberry Plants in 1941.

Treatment.	15.viii.41.		15.ix.41.		8.x.41.	
	Height.	Spread.*	Height.	Spread.*	Height.	Spread.*
Controls	6·5	17·5	8·5	26	5	17
Mild Crinkle Series ..	4·5	22·5	5	13	4·5	12
Severe Crinkle Series ..	3·5	19·5	4	12	3·5	8·5

* One diameter only, at the widest part of each plant.

It will be seen that the vigour of the mild and the severe Crinkle series declined as compared with that of the control series, this being more rapid in the severe Crinkle series. As the control plants were not colonized with aphides at all, this decline may have been due either to the direct effect of the aphis infestation or to infection with Crinkle by the aphides. On the other hand it may have been due to both these factors. That the latter factor was operative follows from the fact that the decline was more rapid in the severe Crinkle than in the mild Crinkle series, the infestation of both series being approximately equal.

As only one diameter (the longest) was measured, the figures do not convey adequately the very striking differences in vigour shown by the three series, particularly as they appeared in October. At this time the spread of the control plants was approximately seventeen inches. That of the two Crinkle-infected series, however, was far less, most of the dwarfed and stunted foliage being concentrated within the circumference of the four inch pot, with an occasional leaf straggling outside it.

Conclusions. The above results show that the strawberry aphis (*C. fragariae*) in its alate viviparous female stage functions as a vector of Crinkle virus to the cultivated strawberry in this country. They also indicate a general correspondence between the relative intensity of the Crinkle symptoms (mild or severe) induced in the aphis-inoculated Royal Sovereign plants and that of the symptoms shown by the corresponding plants from which the virus inoculum was obtained.

DISCUSSION.

The fact that individuals of so many different stages in the life history of the strawberry aphis are capable of transmitting Crinkle is of considerable economic importance. There is little doubt that the wingless forms are responsible for the spread of the virus within any given strawberry bed or field, while the winged forms carry and transmit the virus to plants in other fields in the neighbourhood during the spring migrating period. It has been proved experimentally that the winged form is capable of transmitting Crinkle virus in May, the normal spring migration period (Greenslade, 1941).

Further, in the 1941 experiment, winged aphides previously fed on two Royal Sovereign plants showing mild and severe symptoms of Crinkle, respectively, induced without exception symptoms of corresponding intensity on the healthy Royal Sovereign plants to which they were subsequently transferred. This result confirms the findings in this country of Harris (1937), and Harris and King (1942), that the mild and severe forms of Crinkle are etiologically distinct. Further analysis, however, will clearly be necessary before the precise nature of this distinction from the virological standpoint, can be determined (Harris, 1938).

The importance of the strawberry aphis as a vector of both Crinkle and Yellow-edge has already been emphasized. The necessity of obtaining complete control of the aphis in the field is therefore imperative. This can be achieved by fumigating the plants with nicotine vapour. A detailed report on aphis control by this means has been published recently (Massee and Greenslade, 1941).

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The writer is greatly indebted to Mr. R. V. Harris for reading the MS. and for making several suggestions, especially from the virologist's point of view ; and to Mr. R. M. Greenslade for taking the photographs. Mr. Arthur R. Tidy has been responsible for the cultural aspects of the investigations and his keenness throughout the experiments has proved invaluable.

SUMMARY.

1. The strawberry aphis (*Capitophorus fragariae* Theob.) is shown to be a vector of the Strawberry Crinkle virus in this country.

2. Mild symptoms of Crinkle were induced in healthy plants of the common woodland strawberry (*Fragaria vesca* L.) by transfer and feeding of either adult or immature stages of apterous viviparous female aphides, previously fed on plants of the same species showing Crinkle symptoms of like intensity.

3. Severe and mild symptoms of Crinkle, respectively, were induced in healthy Royal Sovereign plants by colonization with alate viviparous female aphides, previously fed on Royal Sovereign plants showing Crinkle symptoms of corresponding intensity.

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PLATE I.

Fig. 1.

Royal Sovereign strawberry plants to which mild Crinkle has been transmitted by artificial colonization with viruliferous aphides—*Capitophorus fragariae* Theob.

Fig. 2.

Royal Sovereign strawberry plants to which severe Crinkle has been transmitted by artificial colonization with viruliferous aphides—*Capitophorus fragariae* Theob.

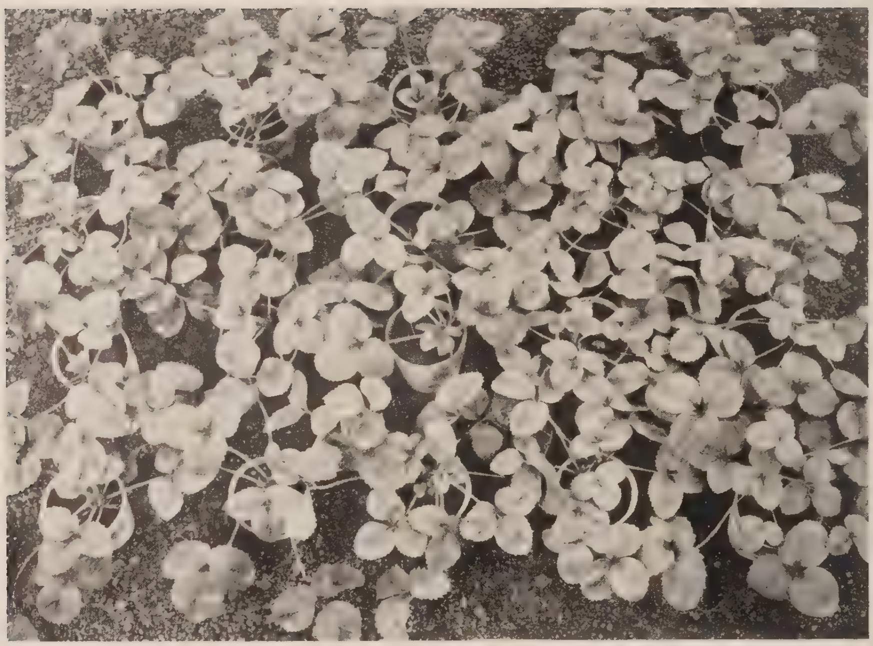

FIG. 3.

Healthy Royal Sovereign strawberry control plants, from the same clone (Malling 40) as those to which the two forms of Crinkle were transmitted by *Capitophorus fragariae* Theob.

THE RELATION BETWEEN MOSAIC INFECTION AND YIELD REDUCTION IN GLASSHOUSE TOMATOES

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INTRODUCTION.

An experiment has already been described (Selman, 1941*a*) in which the effects of certain Mosaic-inducing viruses on the tomato crop were studied. From this work it appeared that crop reduction, due to early infection of the plants, was associated with at least three important effects, viz. (i) reduction in the number of flower buds differentiated, (ii) failure of certain flower buds to set and to mature fruits, and (iii) a slight reduction in the average individual fruit weight. Of the vegetative responses studied, it was noted that the weekly height increment of the main stem was reduced and the weekly rate of appearance of leaves slightly checked by early infection.

The efforts of commercial growers to counteract Mosaic infection by the judicious use of cultural measures frequently yield good results. There is, however, little information concerning the particular primary effect of the virus with which the grower is, or should be, most concerned in his efforts to maintain the weight and quality of fruit picked. An experiment similar to that referred to above, was therefore carried out in 1941, using one strain of tomato Mosaic virus (A.17) only ; and, profiting from the experience gained in earlier work, an attempt was made to overcome some of the known effects of the virus by controlling certain environmental factors.

METHODS AND DESIGN OF EXPERIMENT.

The same glasshouse and soil were used as in the earlier work. Chemical analysis of the soil, sampled to a depth of 6-7 in., after the application of base fertilizers, gave the following results, calculated for soil dried at 100° C. :

Loss on ignition ..	17·94 per cent.	Total P_2O_5	0·779 per cent.
Total Nitrogen ..	0·499 per cent.	Available P_2O_5	0·288 per cent.
Total K_2O	0·493 per cent.	Total carbonates as	
Available K_2O	0·142 per cent.	$CaCO_3$	3·85 per cent.

Comparison of these figures with those of the previous experiment shows that the relative proportions in which the principal elements were present were not the same in the two years, the most important difference probably being in the levels of available potash. Further work is being undertaken to ascertain the relative importance of the fertilizer balance in determining the magnitude of virus-induced crop losses. Both soil analyses, however, would be adjudged satisfactory in relation to the general level of mineral nutrients for a tomato crop.

The experiment was planned to include an investigation of the effect of differential top-dressings of nitrogen on the diseased plants. These were applied during the period April-September, and were as follows :

Medium N_2	Plants received	½ oz./sq. yd. dried blood.
		½ oz./sq. yd. sulphate of ammonia.
High N_2	Plants received	5 oz./sq. yd. dried blood.
		3 oz./sq. yd. sulphate of ammonia.
		plus a mulch of horse manure.

No significant differences could be detected in the flowering and fruiting of the plants grown at the two nitrogen levels, either as between early and late infected plants or within either group. It seems possible that this result may be associated primarily with the rather high level

of total nitrogen initially present in the soil and is not likely to obtain under most commercial nursery conditions.

The house was laid out in a manner similar to that previously described, but was divided into 8 blocks, each consisting of 8 plots of 4 plants per plot, arranged in pairs for testing the nitrogen effects. The positions of the nitrogen twin plots and of the control and infected plots within the twin plots were in random arrangement. The double nitrogen plots were separated by guard rows.

Virus-free seed of the variety Potentate was sown on January 11th, 1941, and the seedlings were transferred to 3 inch pots on February 10th, when the first foliage leaves had appeared. On March 21st, when they were at the 9-10 leaf stage, the plants were transferred to the borders. One half of them were inoculated with tomato Mosaic virus (A.17), in the manner previously described, when the first truss was coming into bloom, on April 3rd. The controls remained healthy until the third week in June, when symptoms of infection began to appear at the time when the fifth truss was blooming. The exact cause of this accidental infection is not known, since elaborate precautions were taken to prevent transmission of the virus when handling the plants. Records were kept of the appearance of symptoms on every plant in the house, and some evidence was obtained which suggested that infection might have occurred in the controls by way of the roots, although exact data concerning the nature and mode of transmission of the virus could not satisfactorily be determined.

Symptom appearance on the inoculated plants began on April 15th and observations and measurements on the flowering, fruiting and growth of the plants were made in the manner outlined in the previous paper. Five flower and fruit trusses were measured separately. Fruit picking started on June 9th and continued until September 4th.

Culture.

One of the most important differences between the experiments of 1940 and 1941 was the improved skill of the author in the later work as a grower of plants. The introduction of this personal factor into scientific work is rightly held to be undesirable. As a key to future objective studies, however, the cautious use of this factor may be of inestimable value in the study of plant growth reactions over relatively long time periods. The indeterminate personal element associated with plant culture must frequently constitute an inherent weakness in much scientific work concerned with the growth of plants where cultural factors have been involved. In agricultural research, the effect of the personal factor has sometimes tended to be subordinated to the effects of meteorological factors in studies of crop plants, since the weather is frequently of paramount importance and meteorological data are readily obtained from nearby stations. With glasshouse crops, however, the environmental factors obtaining within the house are to a very great extent an integral function of the personal skill of the grower. The integration of the minor adjustments of temperature, humidity, and soil moisture, which the experienced grower is able to make intuitively in the culture of his crop, is perhaps the factor which most frequently determines success or failure. The analysis of good " growing " procedure is outside the scope of the present work, but it is certain that average daily or weekly meteorological data, inside or outside the house, can give but little indication of the cultural methods applied, particularly when hourly adjustments have sometimes to be made to house conditions as determined by weather fluctuations. The principles of tomato culture have been dealt with by Bewley (1938), and these have generally been adhered to in this work.

In addition to the improved standard of culture employed, an attempt was made to induce freer flower bud production by increasing the supply of water and nitrogen to the young plants. The results presented below suggest that this attempt was reasonably successful.

Presentation of Results.

The complete data are too bulky to be published in full here, but a copy of them has been deposited in the Natural History Museum, London, S.W.7, for consultation by those interested.

A complete statistical analysis of the results has been made according to the methods of Fisher, using the "z" test of significance. One example of the analysis of variance used is given below. It is proposed to compare the more important results with those of the experiment of 1940 and to assess as far as possible the relative importance of these effects in determining the serious crop losses frequently associated with tomato Mosaic infection.

Yield of Ripe Fruit.

The analysis of variance for the total yields of fruit obtained in 1941 are given below, "z" values judged significant being shown in heavy type.

Analysis of Variance. Yield of Ripe Fruit.

	D.F.	Sums of Squares.	"z" values.
Between plots :			
Blocks	7	1013·0	—
Early *v.* Late Infection	1	2250·0	**1·2310** (1% pt. = 1.0408)
High N. *v.* Medium N.	1	156·0	—
Interaction Virus × N.	1	348·2	0·2976
Deviations (*a*)	21	4029·4	
Within plots :			
Trusses	4	8422·0	**1·3968** (0·1% pt. = 0·7648)
Interaction :			
Trusses × Virus	4	9852·4	**1·4749** (0·1% pt. = 0·7648)
Trusses × N.	4	508·2	—
Deviations (*b*)	116	14960·2	—
Total	159	41539·4	

The level of nitrogen top-dressing was thus found to have no significant effect on the yield of ripe fruit, and no significant effect of this factor could be shown to obtain with the other observations reported below. The importance of truss position was again manifest in this and other variance analyses and indicated the desirability of defining the stage of development of the plant with respect to flower and fruit formation in studies of virus diseases.

In Table I the mean yields obtained from 5 trusses are given, and in Fig. 1 the yields from individual trusses have been plotted graphically for both early and late infected plants in the two seasons. Significant differences are indicated by the heights of the vertical lines to the right of the graphs.

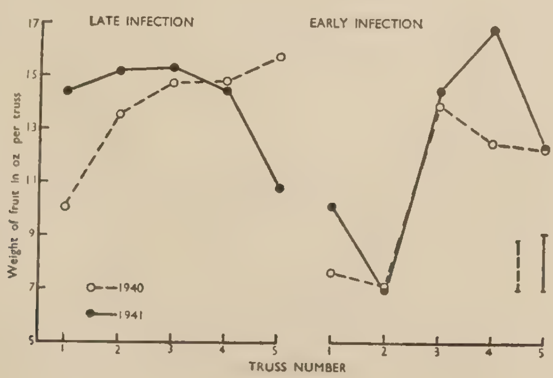

FIG. 1.
Yield of ripe fruit.

TABLE I.

Yield of ripe fruit per plant.

	Late infected (Control).	Early infected.	% reduction due to early infection.
1940	68·8 oz.	53·4 oz.	22·3 Significant.
1941	70·4	61·0	13·4 ,,

The yield of the early infected plants was thus found to be increased in 1941 both absolutely and relatively to the late-infected control plants. Earlier work tended to show that virus-induced yield reduction was associated with effects on flower bud production and the setting of fruit, and, to a less extent, with a reduction in average fruit weight. It will be of interest therefore to see how far these three factors contributed to the improved yield of 1941.

Flower Buds.

An attempt was made to facilitate flower bud production by ensuring adequate supplies of nitrogen and water to the plants when in the 3-inch pots, and later by maintaining the soil of the house at a higher moisture content than was present in 1940. Table II indicates the results of this procedure both on the numbers of flower buds differentiated after inoculation of the early infected plants and on the total numbers of buds produced on five trusses.

TABLE II.

Number of flower buds produced.

	Trusses 2-5.		Total (5 trusses).	
	Control (Late infected).	Early infected.	Control (Late infected).	Early infected.
1940	29·6	26·7	36·2	33·1
1941	37·5	38·5	45·7	46·9

Contrary to the result in 1940, no significant difference was found between the numbers of flower buds produced by trusses 2-5 in the early infected plants as compared with the controls.

Number of mature fruits.

The number of mature fruits per plant was markedly reduced by early infection in both seasons, and this effect was found to be significant in 1941, as indicated by the "z" values given below:

Early *v.* late infection	**1·0329**	(0·7322 = 5 per cent. pt.)
Trusses	**1·5394**	(0·7648 = 0·1 per cent. pt.)
Interaction. Truss × Virus	**1·5632**	,, ,,

The actual numbers of fruits produced by the two groups of plants in the two seasons are given in Table III, and the magnitude of the reduction associated with early infection will be seen to parallel closely that recorded in Table I for Yield of Ripe Fruit.

TABLE III.

Number of mature fruits per plant (5 *trusses*).

	Control (Late infected).	Early infected.	% reduction due to early infection.
1940	30·4	24·0	21·2
1941	29·0	25·8	11·1

Similarly, the numbers of ripe fruits produced on individual trusses parallel closely the fruit yields for those trusses, as shown in Fig. 1. Since no significant differences in average fruit weights could be attributed to the virus effects, this result was only to be expected. The mean truss values for 1941 are shown in Table IV.

TABLE IV.

Numbers of fruits per truss (1941).

	Truss 1	2	3	4	5	Signif. Diff.
Control (Late infected)	6·53	6·33	6·41	5·82	3·95	0·85
Early infected	4·95	2·92	5·98	7·08	4·82	

There was thus a highly significant reduction in the numbers of fruits produced in the second truss of the early infected plants, an effect which was also noted in the earlier work. The second truss was coming into bloom at the time when symptoms of virus infection were appearing on these plants.

Average fruit weight.

In Table V the mean values for average fruit weight are given. A reduction in this value was occasioned by early infection in both seasons, although it could not be demonstrated that the differences were statistically significant in either experiment.

TABLE V.

Average fruit weight.

	Control (Late infected).	Early infected.
1940	2·25 oz.	2·18 oz.
1941	2·46	2·38

Two effects have thus appeared in both experiments in association with the yield reduction occasioned by early infection with tomato Mosaic virus :

(1) A highly significant reduction in the numbers of ripe fruits produced, which is most marked in the truss coming into bloom at the time of appearance of symptoms and which may be independent of the numbers of flower buds produced.

(2) A small, non-significant reduction in average fruit weight.

The effect of early infection on the production of flower buds was eliminated when care was taken to ensure suitable supplies of water and nitrogen to the young plants and to maintain an adequate supply of moisture throughout the growing season.

Blotchy ripening.

A greater percentage of fruit from early infected plants showed blotchy ripening in 1941 than in 1940 (Table VI), although generally the severity of the markings on the fruit was much less serious in the second year.

TABLE VI.

Fruit showing blotchy ripening (percentage by weight).

	Control (Late infected).	Early infected.	
1940	5·4	10·6	Non-signif. difference.
1941	5·6	15·0	Signif. difference.

Fig. 2 illustrates graphically the mean values for the blotchy fruit produced on individual trusses of plants grown in the two seasons. With the early infected plants the percentage of blotchy fruit produced on the second truss was consistently less than the amounts produced on the first and third trusses.

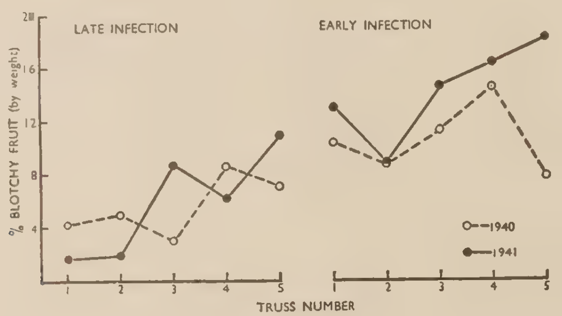

FIG. 2.
Blotchy fruit.

Leaf production and stem growth.

Leaf counts and measurements of the height of the stem were made on 8 plots of both infected and control plants, two marked plants, initially selected at random from each plot, being measured. During the six-week period following inoculation, the controls showed no symptoms of virus infection. The mean values obtained are given in Tables VII and VIII, together with the data obtained for a corresponding period in the previous years.

TABLE VII.

Leaf production.

	Leaves per plant per week (for 6 week period following inoculation).		Total no. of leaves per plant 6 weeks after inoculation.	
	Control.	Infected.	Control.	Infected.
1940	2·72	2·42	29·0	26·0
1941	2·75	2·79	29·4	29·9

The virus-induced reduction in the weekly rate of leaf production that occurred in 1940 was thus not apparent in 1941, neither was any check in the rate of leaf production observed in the second year at the time of appearance of symptoms. Air temperature, which is known to be an important factor influencing the rate of leaf production, was slightly lower in 1941 than in 1940, the means (for 6-week period) being 68·5 ° F. and 72° F. respectively.

Other factors being equal, temperature differences might have been expected to lead to a lower rate of leaf production in 1941 than in 1940.

TABLE VIII.

Height of main stem.

	Inches per plant per week.		Stem height (inches) 6 weeks after inoculation.	
	Control.	Infected.	Control.	Infected.
1940	5·41	4·06	41·4	33·4
1941	4·81	4·59	41·5	40·9

Table VIII indicates that the virus-induced check to stem elongation, noted in 1940 as statistically significant, was almost entirely eliminated in the second season, no significant difference being found between the weekly height increments of the two groups of plants. This result was obtained without detriment to the growth of the controls and was apparently associated with the improved cultural conditions applied, notably higher soil moisture content and greater atmospheric humidity than obtained in the first year.

It appears, therefore, that infection of the plant at the time when the first truss is blooming, does not invariably lead to an interference with either growth rate of the stem or meristematic activity, at least during the six-week period following inoculation.

The results of the quantitative investigations conducted in 1940 and 1941 may conveniently be summarized in tabular form thus :

Effects associated with early infection. Whole plant.

	1940.	1941.
Reduction in total yield of ripe fruit	+ +	+ +
Reduction in number of ripe fruits	+ +	+ +
Reduction in average fruit weight	+	+
Reduction in number of flower buds produced after infection	+ +	–
Reduction in weekly height increment of stem	+ +	+
Reduction in weekly rate of leaf production	+	–
Increase in percentage by weight of blotchy fruit	+	+ +

+ + Statistically significant. + Positive but non-significant. – No effect.

Other disease symptoms.

Leaf symptoms were in most cases confined to a light-green, dark-green mottle, and of the 64 plants inoculated with the virus, not more than 10 showed a yellow-green foliar mottle at any one time. Generally, foliar mottling was less severe in 1941 than in 1940, although air temperatures before May 1st were slightly lower in the second season. Intensity of virus mottling evidently bears no simple relationship to temperature under all conditions. Soil

moisture would appear to be an important factor in controlling symptom intensity in tomato Mosaic disease.

Three plants developed brown stem streaks at the time when foliar symptoms were first appearing on the inoculated plants, together with some leaf necrosis. All three plants rapidly recovered from the initial symptoms and bore fruit.

Enations appeared on the undersides of certain of the leaves of 25 plants during the first week in May. These outgrowths were smaller than in 1940 and relatively inconspicuous.

DISCUSSION.

Following the scheme outlined earlier (Selman, 1941*b*), in which it was suggested that virus infection in its effects on the plant, might, for practical purposes, be regarded as akin to environmental factors, it has been found possible to eliminate certain effects of early infection, previously recorded as statistically significant, by appropriate adjustments of the environment. One serious effect of tomato Mosaic infection has persisted, however, namely the failure of many flower buds to mature and set fruits. Consideration of the problem of fruit set in healthy plants does, however, indicate the important environmental factors which might profitably be investigated in relation to fruit set in infected plants.

Infertile flowers and blossom dropping in the tomato are known to be associated with several factors, the more important being :

(*a*) Deficiency of the elements K, N, B, Mo and Mn. (White, 1938*a* ; Johnston and Fisher, 1930 ; Arnon and Stout, 1939 ; Hoffman, 1933).

(*b*) Environmental factors conducive to a high water-loss/water-uptake ratio of the plant, viz. dry soil, high temperature or low atmospheric humidity (Radspinner, 1922 ; Smith, 1932).

(*c*) Deficiency of carbohydrate. This is a common cause of the failure of glasshouse tomatoes to set fruit in the British Isles during November and December. The short days and low light intensities then prevailing are probably the most important factors in this connection. Two types of plant in which carbohydrate deficiency has been described in association with blossom dropping are recorded by Kraus and Kraybill (1918) in which a low proportion of available carbohydrate to available nitrogen occurred. The influence of the relative availability of carbohydrate and nitrogen on the development of the reproductive organs is incompletely understood. It is of interest, however, to note that Howlett (1936) has shown that carbohydrate deficiency in the tomato induces microspore degeneration and pollen sterility.

Our present knowledge of the effects of tomato Mosaic on plant metabolism indicates that, under certain conditions, infection may lead to conditions within the plant analogous to those resulting from the interaction of environmental factors that induce blossom dropping in the healthy plant.

It is known, for example, that at the time of appearance of symptoms, Mosaic-infected tomato plants are very susceptible to wilting (Selman, 1941*a*). There is a reduction in the percentage water content of all parts of seedling tomato plants infected with tobacco Mosaic virus (Ainsworth and Selman, 1936) and this is most pronounced in the leaves. Such a condition might reasonably be expected to be associated with a water deficit within the plant paralleled by that obtaining in the experiments of Radspinner (1922) and of Smith (1932), and resulting in the failure of fruit to set in the second truss of the inoculated plants of the present experiments. Later, the susceptibility of the plants to wilting has been found to decrease in crop plants ; and in seedlings, the percentage water content increases after an initial fall, eventually reaching a level higher than that of the healthy controls. Such an effect would appear to be paralleled by the partial " recovery " in the fruits set in the third truss of the inoculated plants.

In Mosaic-infected tobacco plants, Cordingley *et al* (1934) have shown that diseased leaves contain lower proportions of carbohydrates than healthy controls. Mosaic-infection of potato is stated to reduce the photosynthetic efficiency of the leaf (Stone, 1936) ; and starch

accumulation in Mosaic-infected tomato leaves (Ainsworth and Selman, 1936) might also be expected to interfere with photosynthesis with a corresponding reduction in the level of available carbohydrates.

Conditions conducive to a high water-loss/water-uptake ratio were suspected of contributing to the incomplete flower setting in the inoculated plants in 1940, accordingly special care was devoted in the second year to ensuring favourable conditions of soil moisture and atmospheric humidity. That little improvement was apparent in the absolute number of fruits set, and the fact that the percentage of flower buds setting fruits was considerably reduced in both control and inoculated plants in 1941, would appear to indicate that moisture conditions are unlikely to be the only factor concerned. Carbohydrate deficiency of Kraus and Kraybill's Type II, or deficiency of some essential elements within the plant, might thus appear to be controlling the set of fruit in the inoculated plants. Deficiency of essential elements in the soil was considered improbable, in view of the heavy manurial dressings applied. An unbalanced fertilizer content of the soil is known to produce unfruitful vegetative plants, similar to those described by Kraus and Kraybill, and it is possible that some such unbalance was effective in accentuating the adverse effects of virus on the metabolism of the plant.

It is of interest to note that the percentage of blotchy fruit was markedly reduced in both seasons in the second truss of the inoculated plants. This was also the truss which produced the smallest number of fruits and the lowest yields. This might indicate that blotchy ripening is associated with deficiencies of essential materials within the plant. White (1938*b*) has stated that blotchy ripening, of the type induced by potash deficiency, is symptomatic of deranged carbohydrate metabolism accompanied by changed water relations of the tissues. It seems possible that the above considerations of the factors relating to setting of the fruit may apply to studies of virus-induced blotchy ripening.

Further work is necessary to ascertain whether all the adverse effects of virus infection can be eliminated by attention to cultural conditions.

SUMMARY.

1. Further data are presented on the effects of inoculating tomato plants growing under glass, with a strain (A.17) of tomato Mosaic virus when the first truss was in bloom, and these are compared with those from uninoculated plants which first showed symptoms of accidental infection when the fifth truss was blooming.

2. Comparison of similar experiments in two successive seasons showed that the following effects of early infection appeared in both :

(*a*) A significant reduction in the number of fruits produced.

(*b*) A small non-significant reduction in average fruit weight.

(*c*) A significant reduction in the yield of ripe fruit.

(*d*) An increase in the percentage by weight of fruit showing blotchy ripening.

3. Contrary to earlier results, early infection induced no reduction in the number of flower buds ; it did not affect the number of leaves produced and had no significant effect on the growth rate of the stem. This result is attributed to improved methods of culture, notably the maintenance of greater atmospheric humidity, higher soil moisture content and an increased supply of nitrogen to the young plants.

4. The importance of the partial failure of the flowers to set fruit in the inoculated plants in relation to final yield is emphasized. At least two causes are believed to be contributory to the failure of the fruit to set :

(*a*) Shortage of available carbohydrate within the plant.

(*b*) A high water-loss/water-uptake ratio in the plant.

5. Leaf enations were produced in both seasons but they were much smaller in size in the second year when moisture conditions were more favourable.

6. The relative reduction in fruit yield due to early infection was 22·3 per cent. in 1940 and 13·4 per cent. in 1941, the absolute yields from both early and late infected plants being higher in the second year.

ACKNOWLEDGMENT.

The author is much indebted to Dr. W. F. Bewley for his advice and encouragement throughout the course of this work. Thanks are also due to Mr. O. B. Orchard for advice on cultural procedure in commercial glasshouses and to Dr. O. Owen for undertaking the analysis of the soil.

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THE NATURE OF THE VOLATILE PRODUCTS FROM APPLES

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INTRODUCTION

It is well known that in addition to the gaseous products of respiration, the apple evolves small quantities of volatile substances. Some of these have a physiological action on the fruit itself and on neighbouring fruits. Ethyl alcohol and acetaldehyde were the first of these products to be identified. A more recently identified one, ethylene (2), has been shown to have a profound influence on certain fruits, causing the onset of ripening changes in them and, in apples, injury to the skin of ripe fruit (3 and 4). Evidence has accumulated of the existence of other physiologically active volatile products of fruit metabolism; for example, the as yet uncharacterized substances which cause Scald in apples (5 and 6).

A preliminary account is given here of attempts to elucidate the nature of certain apple volatile products. Improved methods have been developed for estimating both the ethylene produced by the fruit and the total production of volatile substances in terms of the carbon-content. The use of these methods has shown that ethylene constitutes a high proportion of the total volatile substances produced by the apple.

EXPERIMENTAL

Estimation of total apple volatile products.

In the past a combustion method (7) has been used to measure the volatile products in terms of their carbon-content. The fruit (*ca.* 2 kg.) is placed in a glass desiccator, and is ventilated by an air-stream of 95 per cent. humidity at 3 litres per hour. The carbon dioxide of respiration is removed by a horizontal soda-lime tube (100 cm. by 4 cm. diam.), and the emergent air is led through a combustion furnace, consisting of a silica tube (100 cm. by 1·5 cm. diam.) containing a closely fitting roll of tightly wound copper gauze (30 cm. long) at red heat. Combustion-CO_2 is measured by absorption in standard baryta. This method has the disadvantage that the volatile products are likely to be partially condensed (7) or even chemically modified by the large quantity of soda-lime.

Absorption by sulphuric acid. In an attempt to absorb the volatile products with a view to their subsequent estimation, concentrated sulphuric acid in various forms of gas-washing devices was introduced into the combustion circuit immediately after the desiccator containing the fruit. Discoloration of the acid showed that absorption was taking place in it; but since no marked reduction in combustion-CO_2 resulted, it must be assumed that the substances causing the discoloration were largely removed by the soda-lime when the acid was omitted.

Absorption by activated sulphuric acid. It is well known that the absorptive power of sulphuric acid is greatly increased, especially for ethylene, by the addition of certain metallic sulphates, of which silver sulphate is said to be the most effective (8). The evolution of ethylene from apples is so slight that the suitability of activated acid for the removal of extremely small concentrations of ethylene from an air stream had first to be established. An air stream (3 litres per hour) containing *ca.* 1 part of ethylene in 10,000 parts of air was led through a bubbler and a Truog tower (9), each containing concentrated sulphuric acid, and thence to a combustion furnace. By means of the latter, ethylene that escaped absorption was measured in terms of combustion-CO_2. Concentrated sulphuric acid was ineffective as an absorbent, but when that

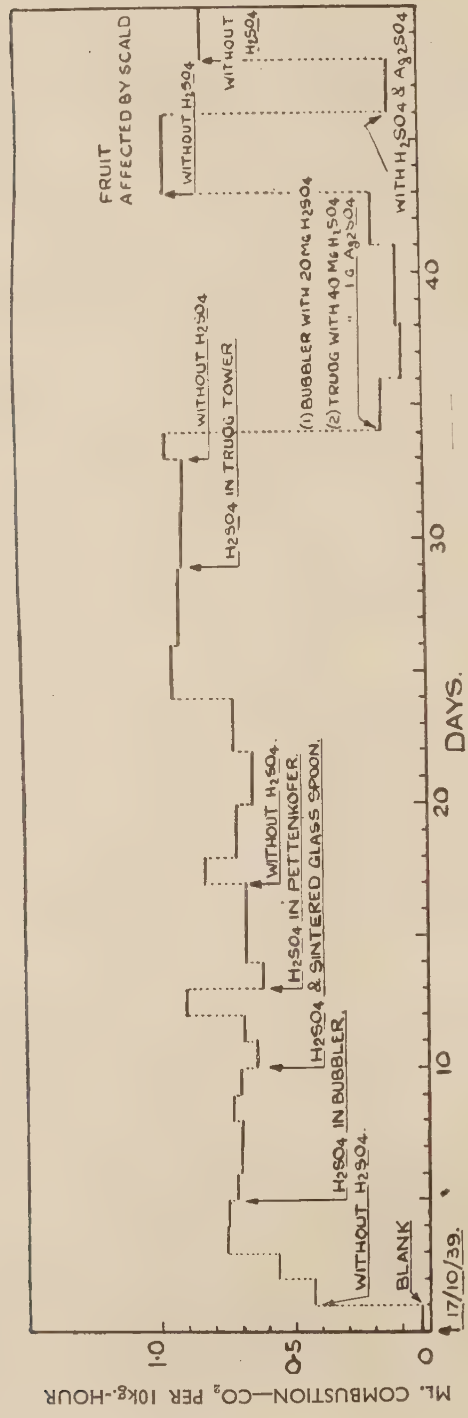

FIG. 1.

Absorption of volatile products from Lane's Prince Albert at 15° C. by sulphuric acid (Respiration *ca* 90 ml. CO_2 per 10 kg. hour.).

in the Truog tower was replaced by activated acid (40 ml. concentrated acid, containing 0·8 g. silver sulphate), over 90 per cent. of the ethylene was absorbed during twelve days, the period of the experiment. The relative absorptive powers of the acids were demonstrated by quantitative oxidation by iodic acid (10) of the untreated acid of the bubbler and the activated acid of the Truog tower : reduction of iodic acid in the former indicated the absorption of less than 2 mg., and in the latter of 63 mg. ethylene. A serious fall in absorptive power occurred when the concentration of silver sulphate fell below 1 g. in 100 ml. acid. It is probable that the formation of a co-ordination complex with the salt is a first stage in the absorption. The possession of a measurable dissociation pressure by this complex would explain the escape from absorption of a small fraction of the gas.

The absorptive capacity of concentrated sulphuric acid for carbon-containing volatile products from apples was next examined. In the combustion-circuit already described, a bubbler containing pure acid, and a Truog tower with an activated acid, were interposed immediately after the desiccator containing the fruit. Absorption was readily measured by the fall in combustion-CO_2. The effect of the addition of mercuric sulphate and of the sulphates of nickel, copper and silver to sulphuric acid, and of silver sulphate to phosphoric acid, was thus investigated. A solution of silver sulphate (2 g.) in concentrated sulphuric acid (100 ml.) proved the most satisfactory (Fig. 1), but the inactivity of the other salts may have been due to their sparing solubility in the acid. Solutions of silver and nickel sulphates together in concentrated sulphuric acid were recommended by Tropsch and Dittrich (10) as being specially effective for the absorption of ethylene, but in the present experiments addition of nickel sulphate conferred no advantage. The efficiency of various sulphuric acid traps in removing volatile products is illustrated in Fig. 1 by their effect on the combustion-CO_2 values as obtained by the furnace method. It is probable that these traps contained the greater part of the apple volatile products, for it is unlikely that any appreciable quantity of them would have passed through the traps and then have been absorbed by the soda-lime scrubbers that followed. Consequently an estimate of the total apple volatile products in terms of their carbon-content was made by applying to each of the traps the method of wet combustion described by Birkinshaw and Raistrick (11). The results are presented in Table I on page 62.

ESTIMATION OF ETHYLENE FROM APPLES.

Several attempts have been made to estimate the ethylene evolved from apples, notably by Nelson and Christensen and their collaborators (12 and 13). The foregoing experiments with sulphuric acid suggested a simple and continuous method. According to Hansen and Christensen (14), "it is improbable that acetylene or olefines other than ethylene are present in apple volatiles". Thus it is likely that in the above experiments ethylene only is absorbed in the Truog tower, the readily condensable volatile products having been removed by the preliminary trap of untreated sulphuric acid. Evidence for this assumption has been obtained. Any ethylene present in the air-stream from the apples should react as follows :

Absorption in activated sulphuric acid :

$H_2SO_4 + C_2H_4 \longrightarrow C_2H_5HSO_4$ (ethylsulphuric acid) .. .. (i)

Dilution and distillation :

$C_2H_5HSO_4 + H_2O \longrightarrow H_2SO_4 + C_2H_5OH$ (ethyl alcohol) .. (ii)

Oxidation by chromic acid :

$C_2H_5OH + O_2 \longrightarrow H_2O + CH_3COOH$ (acetic acid) .. .. (iii)

After absorption (i), the activated acid was diluted with water, and aqueous alcohol distilled off (ii). The latter was oxidized with standard chromic acid (iii) (15), and the amount of oxidizing agent used and of acetic acid produced were estimated by known titration methods. With Laxton's Superb, which gave sufficient volatile products for accurate measurement, the results

TABLE I.

Estimation of total apple volatile products.

Numerical results expressed as ml. CO_2 per 10 kg.-hour from the combustion-CO_2 values (V) in ml., the time in circuit (T) in hours, and the weight of apples (W) in kg., thus $\frac{V \times 10}{T \times W}$.

(1) Sample.	(2) Period of experiment.	(3) Time in circuit.	(4) Wet combustion-CO_2: H_2SO_4 in 1st absorption vessel.	(5) Wet combustion-CO_2: Activated H_2SO_4 in 2nd absorption vessel.	(6) Composition of activated H_2SO_4	(7) Unabsorbed volatile products (in terms of CO_2 of combustion).	(8) Total volatile products (in terms of CO_2 of combustion).	(9) Volatile products by furnace method (in terms of CO_2 of combustion).
Laxton's Superb C_1	24/2/40-5/3/40	239 hrs.	1·46	2·91	35 ml. H_2SO_4 saturated with $NiSO_4$; 0·35 g. Ag_2SO_4	0·42	4·79	3·52
Do. A_2	29/2/40-18/3/40	406 hrs.	1·83	2·90	40 ml. H_2SO_4 with 0·8 g. Ag_2SO_4	0·25	4·98	2·91
Do. B_2	29/2/40-18/3/40	406 hrs.	1·58	2·40	40 ml. H_2SO_4 with 0·4 g. Ag_2SO_4	0·41	4·39	2·68
Do. C_2	9/3/40-18/3/40	218 hrs.	2·14	2·59	35 ml. H_2SO_4 saturated with $NiSO_4$; 0·35 g. Ag_2SO_4	0·42	5·15	3·33
King Edward VII	22/4/40-7/5/40	353 hrs.	0·55	0·83	2·2 ml. H_2SO_4 saturated with $NiSO_4$; 32·8 ml. H_2SO_4 with 0·3 g. Ag_2SO_4	0·21	1·59	1·04

Notes :—

The figures in column (7) record the unabsorbed residuum, which is measured as CO_2 after combustion in the furnace that follows the absorption vessels.

The figures in column (8) are obtained by summation of the values in columns 4 to 7.

The values in column (9) are inserted for comparison and were obtained by making periodic readings of furnace combustion-CO_2 on the same apples in the absence of sulphuric acid traps.

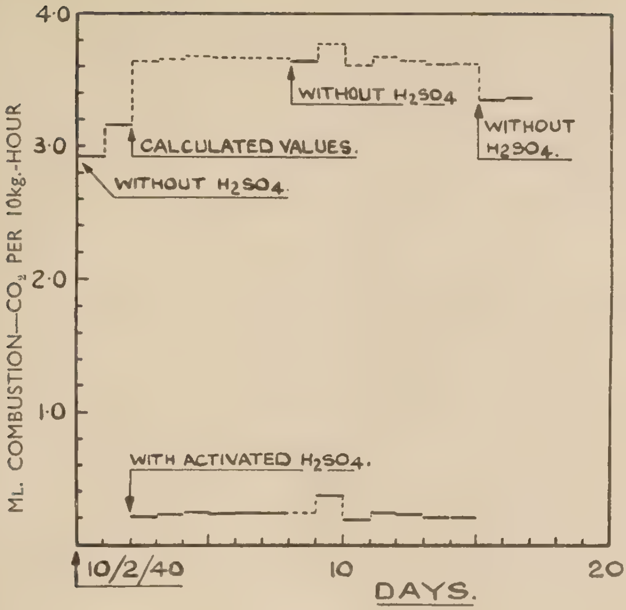

FIG. 2.

Absorption of volatile products from Laxton's Superb at 15° C. by activated sulphuric acid (Respiration *ca.* 120 ml. CO_2 per 10 kg.-hour.

conformed closely with equations (i-iii) (Table II). It is thus reasonable to assume that the volatile product absorbed in the Truog tower was largely ethylene (ethyl alcohol, which would also fit the results, would certainly be absorbed in the preliminary trap (15)). Moreover, the amount of ethylene was sufficient to account for the greater part of the furnace combustion-CO_2, as is illustrated in Fig. 2, in which the upper broken line is calculated from the carbon content of the ethylene trapped in the activated acid and estimated subsequently as ethylsulphuric acid. It agrees well with the results obtained directly from the same circuit in the absence of sulphuric acid traps.

TABLE II.

Estimation of ethylene from apples.

(1) Sample.	(2) Weight of apples.	(3) Time of experiment.	(4) Ethylene absorbed.		(5) CO_2 equiv. to ethylene.	(6) CO_2 per 10 kg.-hour.*
			Calc. from chromic acid.	Calc. from acetic acid.		
Circuit A ..	1,983 g.	288·75 hrs.	124 mg.	123 mg.	387 mg.	3·43 ml.
Circuit B ..	2,114 g.	,,	120 mg.	126 mg.	395 mg.	3·30 ml.

* On the assumption that in the absence of the Truog tower containing activated sulphuric acid the ethylene would be recorded in the furnace method as combustion-CO_2, the figures in column (6) are calculated from the data of columns (2), (3) and (5).

It is probable, therefore, that the wet combustion determinations on the activated acid also give values for ethylene (see Table I). Quantitative oxidation by iodic acid lent further support for this conclusion: the method gives a measure of the oxygen required to oxidize carbon compounds in the traps to carbon dioxide and water. Table III shows that for the activated acid, the carbon-content, as determined by wet-combustion, and the oxygen-requirement, as shown by this method, were generally in fair stoichiometric agreement with the following equation:

$$C_2H_4 \text{ (as } C_2H_5HSO_4) + 3O_2 \longrightarrow 2CO_2 + H_2O \quad .. \quad .. \quad \text{(iv)}$$

TABLE III.

Sample.	Wet combustions.		Iodic acid oxidation.	
	CO_2.	Ethylene equiv. to CO_2.	CO_2 equiv.*	Ethylene equiv. to CO_2.
Laxton's Superb C_1 ..	277 mg.	88 mg.	276 mg.	88 mg.
,, ,, A_2 ..	457 mg.	145 mg.	446 mg.	142 mg.
,, ,, B_2 ..	404 mg.	128 mg.	365 mg.	116 mg.
,, ,, C_2 ..	225 mg.	71 mg.	217 mg.	69 mg.
King Edward	118 mg.	38 mg.	105 mg.	34 mg.

* These values for CO_2 are calculated from oxygen-consumption, using equation (iv).

On the other hand no such correlation was to be expected, nor was it found, between the carbon-content and the oxidation-oxygen of the untreated acid in the preliminary traps, which, as the wet combustion results have shown, had absorbed a considerable amount of carbon-containing volatile products. These are in all probability odorous substances of higher molecular weight and greater condensability, which, it is concluded, are largely, and to some extent reversibly, condensed by the soda-lime scrubbers in the original combustion method.

ADSORPTION EXPERIMENTS.

For the isolation of apple volatile products physical methods are in general unlikely to alter the nature of the substances present. If vapours of odorous substances are present, it would be expected that powerful adsorbents such as activated carbons and silica gels would readily remove them from an air stream. Since the large amount of water vapour produced by apples would quickly vitiate adsorbent properties, a tube of adsorbent carbon (Desorex III) or of silica gel, or of both, was placed in the standard furnace circuit after the soda-lime scrubber, and this largely dehydrated the air-stream leaving the apples. Only a small reduction in the combustion-CO_2 occurred (Fig. 3), a consequence of the fact already established that the volatile products escaping condensation on the soda-lime consist largely of ethylene; for when the tubes were cooled in an alcohol-solid CO_2 mixture to a temperature low enough for the adsorption of a gas such as ethylene, almost complete adsorption occurred (Fig. 4).

Some progress was made towards the identification of the constituents of apple volatile products. Pure air at 15°C. was passed over Lane's Prince Albert apples (35 kg.), contained in a tank, at the rate of 18 litres per hour, and the emergent air was first dehydrated by a calcium chloride tube (100 cm. by 4 cm. diam.) and then led through tubes of silica gel and Desorex III, cooled in alcohol-solid CO_2. Examination of the tubes gave the following results:

(i) *Calcium chloride.* The granules had a strong apple-odour, and the first portion was discoloured. They were dissolved in water, and the solution was steam distilled. In the first runnings a small quantity of a highly odorous oil (A), appeared, and in the later distillates a minute amount of white crystals (B).

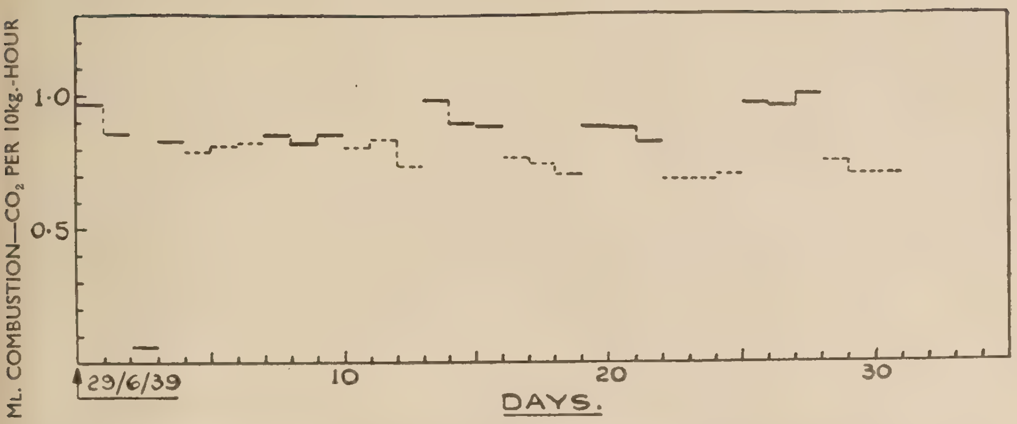

FIG. 3.

Volatile products from Sturmer's Pippin at 15° C. (Respiration *ca* 100 ml. CO_2 per 10 kg.-hour.) The broken lines show results with silica gel at room temperature in circuit.

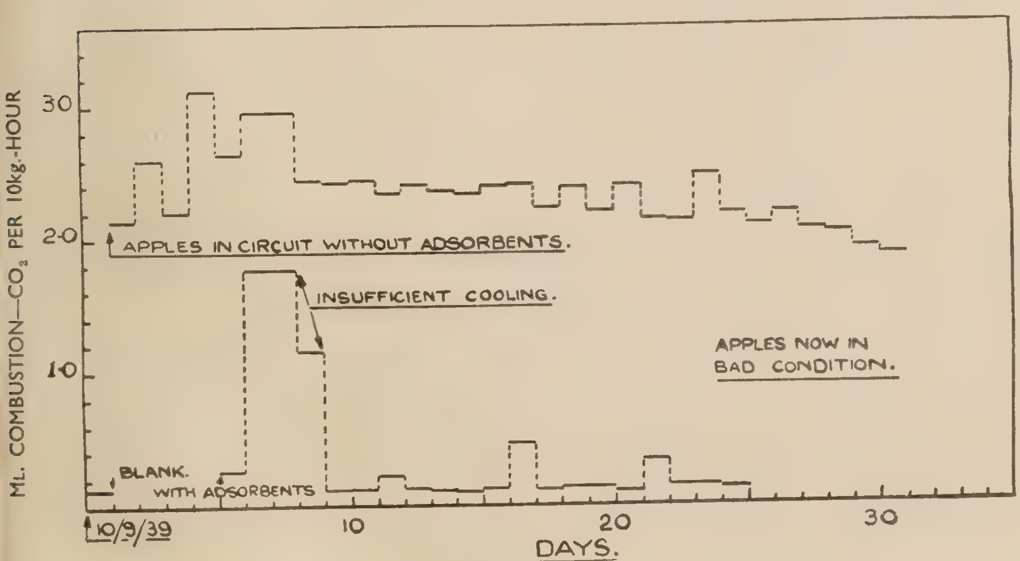

FIG. 4.

Volatile products from James Grieve at 15° C. (Respiration *ca* 100 ml. CO_2 per 10 kg.-hour). The lower values show the effect of adsorption by carbon and silica gel cooled to *ca*—40° C.

(ii) *Silica gel.* The steam distillate here had a strong odour resembling that of musk. The product isolated by ether-extraction was a colourless oil (C) in extremely small yields. Its non-apple odour suggested that one of the original volatile products might have suffered modification in structure on adsorption.

(iii) *Desorex III.* It was expected that the ethylene from the apples would most readily be adsorbed on the carbon, hence, in the steam-distillation, a slow stream of air was led through the apparatus and then through bromine water. An appreciable quantity of ethylene dibromide was obtained, but no other product.

In four similar experiments with calcium chloride (using Lane's Prince Albert, Newton Wonder, Bramley's Seedling and Laxton's Superb apples) fraction (A), subsequently shown to be composed largely of aliphatic esters, was obtained every time, particularly from the non-scalding Laxton's Superb. On the other hand, the less volatile substance (B) appeared in appreciable yield only with Lane's Prince Albert and Newton Wonder, both of them varieties susceptible to Scald. An insufficient quantity was obtained, however, to permit its examination.

The aliphatic esters of apple volatile products.

The product (A) was hydrolysed by aqueous potash to yield an oil having the odour of amyl alcohol; this was separated with ether and condensed with 3 : 5-dinitrobenzoyl chloride in the presence of pyridine (13). The crystalline 3 : 5-dinitrobenzoate thus formed was purified by filtration of its ligroin solution through aluminium oxide and was characterized by its condensation product with α-naphthylamine, which furnished red needles from alcohol, m.p. 79-81° (found by microanalysis: C, 61·4; H, 5·7; $C_{22}H_{23}O_6N_3$ requires C, 62·1; H, 5·4%). The amyl alcohol obtained by Power and Chesnut (1) from the hydrolysis of their steam-distillates of apple parings gave valeric acid on oxidation, and so must have been a primary alcohol. It is probable that the same amyl alcohol was obtained in this case. Of the three primary alcohols, *iso*amyl alcohol (γ-methylbutyl alcohol) and active amyl alcohol (β-methylbutyl alcohol) according to Reichstein (16) give 3 : 5-dinitrobenzoate-α-naphthylamine complexes, melting at a much higher temperature than the product described here. This has been confirmed, a racemic mixture being used for the active alcohol. Moreover, the corresponding derivative of the remaining primary alcohol, *n*-amyl alcohol, has been prepared, and found to have m.p. 87-88°, depressed by admixture with the apple derivative. A possible explanation of the apparent non-identity of the apple alcohol with these is that the former may be an optical antipode of the active alcohol, the derivatives of which, by analogy with similar types, may melt at lower temperatures than the racemate. It is hoped to obtain further information on the identity of the alcohol, when sufficient material becomes available.

The aqueous alkaline residue from hydrolysis was acidified with phosphoric acid and then distilled. The distillate, which smelt of acetic and higher aliphatic acids, reduced silver nitrate. It changed the colour of alkaline potassium permanganate first to blue and then to deep green, with final precipitation of manganese dioxide. These reactions are characteristic of *formic acid*. Neutralization of the distillate with chalk, followed by evaporation and dry distillation, yielded acetone, as indicated by a strong indigo reaction with an alkaline solution of *o*-nitrobenzaldehyde. Under these conditions this is a specific reaction for *acetic acid*. Owing to the small quantity of material available, it was not possible to identify other acids.

SUMMARY.

It has been found that whereas concentrated sulphuric acid will not absorb apple volatile products completely, an acid activated by the addition of silver sulphate is strikingly effective. The almost complete absorption thus made possible offers a means of estimating the total volatile products in terms of carbon dioxide. The condensable and odorous constituents were absorbed in a preliminary trap of ordinary concentrated sulphuric acid, and wet combustions

indicated that about one-third of the volatile products was thus removed, a proportion that tended to increase with over-ripeness of the fruit. A second trap, containing activated acid, accounted for most of the remaining two-thirds. The addition of silver sulphate to sulphuric acid has a like effect on similar concentrations of ethylene in air under analogous conditions. Moreover, the activated acid from the volatile products-circuit reacted quantitatively with chromic acid in a manner that could be explained only on the assumption that the absorbed substance was ethylene. It is concluded, therefore, that ethylene forms a high proportion of the total volatile substances produced by the apple over a long period of its storage life. About the same proportion occurs for a variety that gives much ethylene (e.g. Laxton's Superb) as for one that gives but little (e.g. King Edward VII), a rather surprising result, since different mechanisms might have been attributed to the production of ethylene and of other volatile substances. Owing to condensation on the soda-lime scrubber, the method of estimation of volatile products by furnace-combustion gives low values.

Considerable evidence has accumulated during many years, particularly in work on the nature of Scald, of the production by fruit of volatile substances more readily condensable than ethylene, and at least in part odorous. This has been confirmed by the separation from several adsorbents of a variety of odorous substances, among them esters of amyl alcohol with formic and acetic acids.

The work described above was carried out as part of the programme of the Food Investigation Board, and is published by permission of the Department of Scientific and Industrial Research. Technical assistance was given by Mr. J. North.

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BOOK REVIEWS

MODERN FRUIT PRODUCTION. By J. H. GOURLEY and F. S. HOWLETT. (New York, The Macmillan Co. ; London, Macmillan & Co., 1941, pp. vii and 579, 87 figs., 18s.)

This is the kind of book that most progressive fruit growers, and certainly all teachers of scientific horticulture, have for so long hoped would be written by a British author. It possesses that happy blend of science and practice which has so often been aimed at in recent larger horticultural books and so rarely achieved. Not overloaded with experimental details it yet presents modern scientific theory in its true relation to practical method. With only an elementary knowledge of chemistry and botany the student can follow the arguments without difficulty, while the practical details are full and complete.

Although the book is written for American conditions it is far less parochial in outlook than most of its kind, and it is certainly refreshing to find in it many references to British publications.

The arrangement of the matter is somewhat illogical ; the subject of factors affecting flower formation (Chapter 3) is dealt with before choice of site and planting, this being followed by cultural practices, fertilizers and pruning, with which flower formation is so closely connected. Similarly, winter injury, nutrient deficiencies, physiological disorders, propagation and stocks are left to the end, after storage of fruit. They would better have been dealt with earlier in the volume.

The book is, however, one that can be recommended to all students of scientific fruit production as probably the best of its kind that has yet been produced.

R.H.S.

IMPERIAL BUREAU OF HORTICULTURE AND PLANTATION CROPS. Index to *Horticultural Abstracts*, Volumes I-X, 1931-1940. I.B.H.P.C., East Malling, Kent, 1941, pp. 160, 25s. net.

After ten years, the value of the series of abstracting publications by the various Imperial Bureaux has been proved conclusively. As a ready means of keeping up to date with world-wide literature and as a source of reference these abstracting publications have proved to be of inestimable importance to research workers and teachers. *Horticultural Abstracts*, since its first appearance in 1931, has been an outstanding example of the series. The subject covers so wide a field in itself and has so many connections with the applied and pure sciences that the preparation of this publication has entailed a vast amount of examination and critical selection of the literature, work most faithfully and carefully carried out by the Bureau. Its text and indexes provide a most complete annotated bibliography of horticultural research literature published in every part of the world since 1930.

The value of *Horticultural Abstracts* has now been very greatly enhanced by the preparation of a comprehensive subject and author index to the first ten volumes. The practice of issuing such cumulative indexes is fortunately increasing, but few approach this one in completeness of cross-references. When it is realized that more than 9,500 papers have been abstracted in the ten volumes, and that each is referred to in the accumulated Index under some three, four, or more headings, the magnitude of the task so successfully accomplished by Dr. Akenhead, Deputy Director of the Bureau, can be apprehended. The Abstracts, with the addition of this Index, now become a complete and easily used guide to all the literature likely to concern the horticultural research worker since 1931.

The only criticism that can be levelled at the publication is its price. Twenty-five shillings is a great deal to pay for what is, after all, only an index of no use without the Abstracts. The Agricultural Research Council would have spent money well by subsidizing such a volume to bring it within the reach of everyone subscribing to *Horticultural Abstracts*.

R.H.S.

A PROMISING ATTEMPT TO CURE CHLOROSIS, DUE TO MANGANESE DEFICIENCY, IN A COMMERCIAL CHERRY ORCHARD

By J. B. DUGGAN*

Bucks. War Agricultural Executive Committee

INTRODUCTION.

It is not unusual to find an orchard in which the trees—especially their leaves—are obviously unhealthy, in spite of sound management and satisfactory manuring. Diagnosis of the cause has often proved difficult, yet it is most unsatisfactory both to adviser and grower to leave such cases unsolved. The present one, although concerned with manganese deficiency alone, indicates that methods of diagnosis similar to those described here might be used with advantage where deficiencies of other elements are suspected.

During an advisory visit in 1937 it was noticed that the larger part of a certain cherry orchard was in good heart and bore good crops, but that a portion of it, containing some 100 trees, had ceased to be profitable, while the worst affected trees bore no fruit.

SYMPTOMS OF THE MALADY.

The earliest easily detectable symptom was a yellowing of the leaf, beginning along the margin (Plate I, Fig. 1) and spreading between the veins. It increased in intensity and extent until only the midrib and the bases of the lateral veins remained green (Plate I, Fig. 1, b, c and d). This was followed by the progressive appearance of a brownish-purple colour in the marginal interveinal areas, more conspicuous where the leaf was widest and most marked on the undersurface. Finally, scorching started along the margin and spread, the midrib and bases of lateral veins being the last to become affected (Plate I, Fig. 1, e). Some of the leaves became completely yellow, but the midrib and bases of the lateral veins usually remained green, even after the beginning of marginal necrosis.

The affected leaves, especially those on the spurs, were much reduced in size. New shoot growth was reduced according to the severity of the chlorosis. A healthy cherry tree makes an annual new shoot growth of 9 in. to 12 in., but in the affected trees this was often less than 1 in. Later, as chlorosis grew worse, first the spurs and later quite large boughs died, but in no case had a whole tree died as a result of the trouble.

With trees not seriously chlorotic there was a marked difference between the spur leaves and those on the young growing shoots ; the former were chlorotic, the latter frequently green. This was particularly noticeable on the umbrella-shaped variety Governor Wood. In it, the new growth from the rounded head was frequently green, in marked contrast to the yellow colour of the spur leaves. The reduction of crop was in direct proportion to the severity of the chlorosis. In trees only slightly affected the quality of the fruit was much reduced ; it was more highly coloured, harder in texture, less juicy and smaller than normal. The effect was very evident even before the leaves showed marked symptoms, and trees showing only mild symptoms rarely produced a heavy crop.

VARIETIES AFFECTED.

Different varieties were unequally affected. Thus, trees of Governor Wood were very small, some were nearly dead, and all exhibited advanced stages of the disorder. Amber (syn. Biggareau Kentish) and Roundel Heart were less affected, whilst Early Rivers and Biggareau Napoleon showed only the first symptoms. These differences were in part due to soil variation but may have also been partly due to varietal differences of response.

* Work carried out when the author was Assistant Adviser in Commercial Fruit Growing to the County Adviser, and on the staff of the Kent Farm Institute, Borden, Sittingbourne.

Several other cherry orchards in the neighbourhood seemed to be affected by the same, or a similar trouble. One was as severely affected as the one under consideration, and resembled it in soil condition and situation. In some, the trees were interplanted with pears, plums and apples; none of these appeared to be as severely affected as the cherries. Mild symptoms were observed on apple leaves but none on pears and plums.

Preliminary diagnostic work.

Spectrographic analysis.

From the symptoms it appeared that the affected trees were suffering from a deficiency of one or more elements necessary for healthy growth. Samples, each consisting of the top two fully developed leaves from each of ten current season's growths, were taken from healthy and unhealthy trees and were sent to Mr. S. G. Thompson at East Malling for spectrographic analysis. It was found that the spectrogram of the unhealthy sample gave a manganese line considerably less dense than that of the healthy one, suggesting serious manganese deficiency. The only other abnormal feature was a magnesium line slightly less dense than normal.

Physical character of the soil.

In the orchard concerned there was a marked difference between the soil bearing the chlorotic trees and that bearing the healthy ones, the boundary between them being sharply defined. The former were on a shallow, eroded soil, derived from the Thanet Beds, showing signs of bad drainage, while the latter were on a deep Valley Brickearth, the drainage of which was mainly quite good. The Thanet Beds outcropped on the side of a small valley, and erosion had caused the soil to be shallow. It consisted of about 10 in. of greyish-brown heavy loam (very fine sandy clay-loam), showing slight signs of mottling due to seasonal water-logging. Below this lay the Thanet Beds, orange, yellow and pale grey mottled. In some places the texture remained the same as that of the surface soil, while in others it stiffened to a very fine sandy clay. The soil of the Valley Brickearth, on the flatter land, consisted of a light greyish-brown loam at the surface (very fine sandy loam) passing almost imperceptibly into the typical warm brown of the underlying Valley Brickearth. The texture stiffened below until at about 2 ft. the soil became a heavy loam (very fine sandy clay-loam) ; but below this it lightened once more at 3 ft. 6 in., to loamy, very fine, sand. There were only the faintest signs of seasonal waterlogging in the soil, and these were all below 2 ft. The down-wash origin of the Valley Brickearth was shown by the presence of small fragments of chalk throughout the whole depth of the soil.

Experience gained from soil surveys has shown that cherry trees are more susceptible to adverse soil conditions than most fruit crops (2). Their main soil requirements are adequate depth, good drainage and medium texture. The shallow, poorly drained, heavy loam of the Thanet Beds complies with none of these conditions and thus could reasonably be regarded as unsuitable for cherry growing. The main element of surprise lay in the fact that some varieties had made quite vigorous growth and had reached a considerable size on that soil. The Brick-earth soil, on the other hand, complied with the requirements of the cherry tree in all respects. The drainage deficiency was, indeed, very slight, and the depth of well drained soil above, was sufficient to produce a good tree.

Examination showed that the main difference between the two types of soil was the depth of the well-drained portion. The pH values were the same, $7 \cdot 7$ from the 0″-8″ horizon, in one case associated with free and in the other with impeded drainage. This value approaches the point where lime-induced chlorosis (deficiency of iron in the tree) might be expected. On the other hand, manganese deficiency is commonly shown by susceptible plants under conditions of high pH and poor drainage, and this was already suggested by the results of the spectrographic analysis. Thus, the evidence pointed first to a probable deficiency in the unhealthy trees of manganese then iron and possibly magnesium.

To check the preliminary diagnosis, solutions of Mn, Fe and Mg salts were injected into the unhealthy trees, first on a small scale and later on a more extensive one.

1. *Small scale diagnostic injections.*

Roach's methods of shoot-tip (4) and leaf-stalk (4) injection were used. The latter was more suitable than the former, because sheep in the orchard interfered with the bent down shoot-tips immersed in the solutions employed.

The salts used were ferrous, manganese and magnesium sulphates, each at a concentration of 0·025 per cent. in distilled water.

Shoot-tips and leaf stalks on three trees were injected on July 1st, 1938, each injection being replicated a number of times. The results visible after 17 and 30 days, respectively, are set out in Table I.

TABLE I.

Shoot-tip and leaf stalk injection results.

		18 vii. 1938.	31 vii. 1938.
Ferrous sulphate	1	No response	No response
	2	,, ,,	,, ,,
	3	,, ,,	Slight improvement
	4	,, ,,	No response
	5	,, ,,	,, ,,
	6	,, ,,	,, ,,
Manganese sulphate	1	Slight improvement	Marked improvement
	2	,, ,,	,, ,,
	3	No response	No response
	4	,, ,,	Very marked improvement
	5	,, ,,	,, ,, ,,
	6	,, ,,	,, ,, ,,
Magnesium sulphate	1	Very slight response	No response
	2	No response	,, ,,
	3	,, ,,	,, ,,
	4	,, ,,	,, ,,

It will be seen that by July 31st five out of six shoots injected with manganese sulphate showed a response; only one shoot was slightly improved by the iron salt and there was practically no response to any of the magnesium sulphate injections.

2. *Branch injections.*

Injections were also made into some of the smaller branches of a Governor Wood tree. The branches were about 10 ft. long and well provided with fruit spurs. The trees had made

TABLE II.

Results of small branch injection.

	Amount of solution taken up.	29 ix. 1938.	27 vi. 1939.	30 vi. 1940.
Ferrous sulphate	250 cc.	No response	No response	No response
Manganese sulphate	500 cc.	No response	Leaves completely green	Leaves a full dark green
Manganese sulphate + Ferrous sulphate ..	500 cc.	No response	Leaves completely green	Leaves a full dark green

only a negligible amount of growth for several years and the leaves were markedly chlorotic. Suitable boughs growing alongside the injected ones served as controls. The injections were made 6 ft. from the tip of each branch on July 31st, the solutions being of the same concentration as before. The results obtained in the same and in two successive years are given in Table II.

It is interesting to note that the effect of injecting manganese sulphate was not apparent in the first year. Positive results from injections at the end of August, 1938, did not show up until the summer of 1939. There was no response to the injection of iron, but the mixture of manganese and iron salts was as good as, but not better than, manganese sulphate alone.

Chemical analysis of the soils.

The effects of the injections were so clear cut as to leave little doubt that the trees were deficient in manganese, and this seemed to justify chemical analysis of the soil. Samples were, therefore, taken with an auger from three horizons around healthy and unhealthy trees in July, 1939. The analytical results are given in Table III.

TABLE III.

Results of chemical analysis of soil.

Horizon. (inches.)	Available potassium. (12.)	Available phosphorus. (12.)	Organic matter.	pH.	Total manganese. (mg. per 100 gm.)	Available manganese. (mg. per 100 gm.)
	Deep Valley Brickearth around healthy trees.					
0–8	+ extra high	high	3·63	7·7	29·3	2·09
8–16	extra high	medium high	1·11	7·7	27·5	0·88
16–28	low-medium	medium-medium high	0·81	7·6	33·1	0·74
	Shallow eroded Thanet Beds around unhealthy trees.					
0–8	+ extra high	high	3·73	7·7	26·6	1·06
8–16	extra high	medium high	0·74	7·6	13·1	0·32
16–28	high	medium high	0·64	7·6	—	—

The figures show that there is no deficiency of available potassium or phosphorus in the soil around either healthy or chlorotic trees. The amount of available potassium is extremely high in both instances, and there is more in the top 28 in. of the soil under the chlorotic trees. It is possible that such high amounts reduce the uptake of other nutrients, e.g. manganese, and this effect is more pronounced in shallow, badly-drained soil.

Under both healthy and chlorotic trees the quantity of organic matter in the soil is relatively low and of the same order, and alkalinity is the same. It has been observed in other instances (Wain, 13) that when a soil is alkaline and badly drained, symptoms of manganese deficiency commonly appear in susceptible arable crops, and the deficiency is more pronounced when the amount of organic matter is high. Roach (6), on the other hand, has found an association between low organic matter and deficiencies of iron, manganese and zinc. The cause is not known, but the result may be due either to the formation of manganese compounds of low availability or to lowered uptake of available manganese by the roots. The formation of compounds of low availability may be determined by alkalinity, by alternate oxidation and reduction and by the nature of the organic matter. Lowered uptake of available manganese may be due to an excess of soluble calcium salts in the soil solution, and this is more likely to arise when water moves slowly through a calcareous soil than when it moves freely. Morley Davies (3) reports Lundegårdh's data showing reduced amounts of manganese in the leaves of sugar beet with increase in the amount of calcium in the soil.

There is less total manganese in the soil under the chlorotic trees, particularly in the second horizon, but in both areas the amounts are not high. The amounts of available manganese,

however, are very high when compared with those found in soils where manganese deficiency of arable crops occurs ; but the amount in the top 16 in. of soil under the chlorotic trees is less than half the amount found in the soil under the healthy ones. Two possibilities arise : either cherries require high amounts of available manganese, or there are factors reducing the uptake of available manganese by the chlorotic trees. The samples were taken from both areas at the same time, because the available manganese in the soil probably fluctuates in amount throughout the year.

ATTEMPTS TO REMEDY MANGANESE DEFICIENCY.

It is often supposed that deficiencies of minor elements in the soil may be remedied by applying dung, and Morley Davies (3) reports a considerable reduction of manganese deficiency in arable crops as a result of incorporating farmyard manure. A small test of the effect of applications of heavy dressings of farmyard manure was therefore set up. It was considered that adding a soluble manganese salt to the soil would have little effect on the unhealthy trees, as the manganese would rapidly become unavailable. Reduction of pH by adding an acid or sulphur to the soil was considered too expensive, and impracticable in view of the amount of free chalk present in it.

1. *Manurial treatments.*

Three equally chlorotic trees were selected for each treatment. Their leaves were mainly yellow except for the midrib and the bases of the lateral veins ; growth was negligible and cropping very poor.

Roach (7) has shown that treatments applied to the soil under only half of a tree affect only those branches immediately above the treated side, though some allowance must be made for a slight twist in the trunk. Thus, by treating one side of a tree the other side can be used as a control. This method of procedure was adopted in the present trials.

Treatment A.—The soil under half the tree was dug as deeply as the roots would allow, to a distance of 3 ft. beyond the spread of the branches.

Treatment B.—As for *A*, but with the addition of farmyard manure over the dug surface, at the rate of 55 tons per acre.

Treatment C.—The soil was not dug ; farmyard manure, at the same rate as in *B*, was spread over the grass.

The treatments were carried out during the winter of 1937-38. During the summers of 1938 and 1939 close observation of the trees was made from time to time but no appreciable difference was ever observed between the treated and untreated sides of the trees. It was therefore concluded that neither cultivation nor the application of farmyard manure would cause any improvement in the trees.

2. *Spraying.*

With certain other crops a deficiency of manganese has been made good by spraying with a manganese lime spray (Thompson, 11) or with a solution of manganese sulphate, although no permanent effect has been observed (Wallace, 15). Marsh Spot in peas has been prevented in Holland by spraying pea crops with manganese sulphate solution at about flowering stage. Spraying cherries two or three times a year is not difficult provided the applications do not coincide in time with fruit picking. Moreover, spraying machinery is usually available on farms where cherries are grown.

Manganese sulphate was used at concentrations of 0·25%, 0·5%, 0·75% in distilled water. Each treatment was given with and without a spreader, to ascertain whether the thinner and more even coating of spray due to the spreader would increase the intake of manganese by the leaves. A manganese lime spray fluid (made by adding 1 ounce of calcium hydroxide to 2½ gallons of water and adding 2 ounces of manganese sulphate to this mixture) was included.

Four-foot lengths of horizontal branches were selected on trees showing severe chlorosis. Suitable controls were labelled alongside each treated branch. Each treatment was duplicated. Spraying was carried out on a calm day in July, 1939. The evening was chosen to allow the spray to remain in a liquid condition for as long as possible.

Observations of the sprayed branches were made throughout 1939 and 1940 and the results are presented in Table IV.

TABLE IV.

Results of spraying tests.

Treatment.	Results.		
	4 viii. 1939.	20 vi. 1940.	20 vi. 1941.
A.1. Manganese sulphate 0·25%	1. Slight improvement over control	Improvement maintained	Improvement not maintained
,,	2. ,,	,,	,,
Manganese sulphate 0·25% + spreader*	3. No improvement	No improvement	No improvement
,,	4. ,,	,,	,,
B.1. Manganese sulphate 0·5%	5. New leaves greener, but old leaves no change	Slight improvement over control	,,
,,	6. No change	No change	,,
2. Manganese sulphate 0·5% + spreader*	7. Slight improvement in green	Improvement still evident	,,
,,	8. New leaves greener but old leaves no change	,,	,,
C.1. Manganese sulphate 0·75%	9. No change	No change	,,
,,	10. Slight improvement in green	Improvement still evident	,,
2. Manganese sulphate 0·75% + spreader*	11. No change	No change	,,
,,	12. New leaves greener than control	,,	,,
D.1. Manganese sulphate + lime	13. Appreciable improvement in green colour	Slight improvement over control	,,
,,	14. ,,	,,	,,

* Sulphonated lorol 0·05%.

No damage was observed from any treatment at any time. Each treatment was followed by some improvement, which increased with increasing concentration, but no improvement was observed when a spreader was added. The best result was obtained from manganese sulphate + lime spray. In no case was the improvement so well marked as to give promise that one or two applications would provide a commercial cure for the deficiency. This does not rule out the possibility that, by increasing the number of applications and the concentration of the spray fluid, a cure might be possible ; but the results were not promising.

3. *Liquid injections into large branches and whole trees.*

As injections into small branches had been completely successful in 1938, the next step was to inject large branches and whole trees with manganese sulphate solution. At the same

time it was thought expedient to inject with ferrous sulphate also, since in 1938 only one small branch had been injected with it and the results were negative. The trees and branches for injection were selected as being as uniformly chlorotic as possible. Growth had ceased, cropping was very poor and the leaves were yellow, with the midribs and bases of the lateral veins green. Roach's (4) methods were used, and the first injections into large branches were made on June 27th, 1939. Table V summarizes the treatments and the responses obtained.

TABLE V.

Results of branch injections.

Treatment.	Diameter of branch.	Amount of sol. taken up and time taken.	Observations.		
			4 viii. 1939.	30 vi. 1940.	23 x. 1941.
SMALL BRANCH. Manganese sulph. 0·025% sol.	$2\frac{1}{2}''$	9 litres in 6 days	No change	Complete return of green in the leaves	Leaves still green standing out conspicuously from rest of tree
LARGE BRANCHES. Ferrous sulph. 0·025% sol.	7″	,,	,,	No change	No change
Manganese sulph. 0·025% sol.	8″	,,	,,	Some evidence of better green	No better than control branches
Ferrous sulph. 0·025% sol.	7″	,,	,,	No change	No change

In the attempts to inject large branches the problem was to get sufficient quantities of solution absorbed. In all cases, 9 litres of solution took not less than six days to pass into the branch. Compared with other fruit trees this was most unusual, for an apple branch of comparable diameter to branches 2 and 3 will absorb 18 litres in thirty-six hours. At no time was any damage observed due to absorption and it was inferred that much greater quantities of solution than those actually taken up would be necessary to cause the return of the green colour to the leaves and ensure its persistence for several years.

As weather conditions and time of year might have influenced the intake, further injections were made on July 31st. In order to increase the amount of salt taken in, the concentration of

TABLE VI.

Results of large branch and whole tree injections.

Treatment.	Diameter of trunk or branch.	Amount of sol. taken up and time taken.	Response.	
			4 viii. 1939.	30 vi. 1940.
Ferrous sulphate 0·05% ..	6 in.	9 litres, 4 days	None	None
Manganese sulphate 0·05%	7 in.	4·5 litres, 4 days	Leaves slightly greener than control	Definite improvement, but not yet normal
Manganese sulphate 0·05%	6 in.	2·5 litres, 4 days 6·75 litres, 11 days	None	Much greener than control, not yet normal
Ferrous sulphate 0·05% ..	4·5 in.	9 litres, 4 days	None	None

the solutions used was doubled. The rate of absorption remained slow as will be seen from Table VI.

It should be explained that injection of the small branches was through a $\frac{3}{16}$ in. diameter hole, drilled transversely right through the centre of the branch. The injections into larger branches were through two or more holes, each $\frac{1}{4}$ in. diameter and 2 in. to 3 in. deep. During injection, intake of the solution by larger branches frequently stopped and started again only after reboring the hole slightly larger, or by making a fresh hole. Various sizes of holes were tried, up to $\frac{1}{2}$ in. diameter ; the larger holes increased the rate of intake, but it was still very slow. No satisfactory explanation can be given of the difference in behaviour between small and large branches.

The above results show that injection of solutions into cherry trees in quantities sufficient to effect a lasting cure of chlorosis is not yet practicable.

4. *Solid injection of large branches.*

The method of solid injection was adapted from that described by Bennett (1). This consists of the injection of the dry salt into holes bored in the trunk or branches of the tree, the amount of the salt depending upon the diameter. In Table VII, which is adapted from Bennett's table, the appropriate amount of $MnSO_4$ for a given diameter is stated.

TABLE VII.

Dosage and hole sizes for injection of dry manganese sulphate.

Tree diameter.	Holes.			Amount of manganese sulphate.	
	Number.	Diameter.	Depth.*	per hole.	per tree.
inches.		inches.	inches.	grammes.	grammes.
1	1	$\frac{1}{4}$	$\frac{5}{8}$	0·25	0·25
2	1	$\frac{1}{4}$	1	·1·00	1·00
3	3	$\frac{1}{2}$	$1\frac{1}{2}$	0·75	2·25
4	4	$\frac{1}{2}$	$1\frac{1}{2}$	1·00	4·00
5	5	$\frac{1}{2}$	$1\frac{1}{2}$	1·25	6·25
6	6	$\frac{1}{2}$	2	1·50	9·00
7	7	$\frac{1}{2}$	2	1·75	12·25
8	8	$\frac{1}{2}$	2	2·00	16·00
9	9	$\frac{1}{2}$	$2\frac{1}{2}$	2·25	20·25
10	10	$\frac{1}{2}$	$2\frac{1}{2}$	2·50	25·00
12	12	$\frac{1}{2}$	$2\frac{1}{2}$	3·00	36·00
14	14	$\frac{1}{2}$	3	3·50	49·00
15	15	$\frac{1}{2}$	3	3·65	55·00
20	20	$\frac{1}{2}$	3	5·00	100·00

* Depth of hole varies with the thickness of the bark and the thickness of the cork used for sealing the hole.

Manganese sulphate and iron citrate, in solid form, were separately injected with a special glass gun into holes $\frac{7}{16}$ in. diameter, having a depth depending upon the diameter of the branch. Each hole was afterwards sealed with a tightly fitting thin piece of cork, pressed to a depth just below the level of the bark to form a cushion over which the callus tissue grew. The hole was finally sealed with adhesive tape.

Three large, poorly growing branches, carrying badly chlorotic leaves, selected in the previous autumn when the symptoms were clear, were thus injected on March 24th, 1939. The results observed during 1939, 1940 and 1941 are given in Table VIII, and the effect of the injections on foliage colour and cropping can be seen in Plate I, Fig. 2.

These results show that injections of solid manganese sulphate completely cured the chlorosis, and the cure lasted for three growing seasons. No signs of the disease have been evident at any time since injection. The green colour of the leaves bore comparison with that

TABLE VIII.

Results of solid injections into branches.

Treatment.	August, 1939.	July, 1940.	July, 1941.	July, 1942.
Manganese sulphate Branch 5″ diam.	Complete return of leaf to a good green colour. Growth in excess of control	As for 1939	As for 1940 but branch heavily laden with fruit. Control very few fruits	Green colour of leaves maintained
Manganese sulphate Branch 6″ diam.	,,	,,	,,	,,
Ferric citrate Branch 6″ diam.	No response	No response	No response	No response

of the leaves of the healthiest trees in the orchard. The failure of the iron injection in this and the previous trials showed that it was not worth while proceeding with this element and the next thing was to try the injection of solid manganese sulphate into whole trees.

5. *Solid injection of whole trees in* 1941.

Twenty trees showing severe chlorosis were marked in the autumn 1940 for injection in the spring of 1941. It was thought worth while to try whether smaller doses than those used in 1940 would suffice to produce equally good results ; hence in addition to the full one (following Bennett) half- and one-seventh doses were used. Moreover, to save labour and damage to the trees, the number of holes was reduced in some trees from one hole per 1 in. of the trunk diameter to one hole for each $1\frac{1}{3}$ in. of the diameter. The amount of the salt per hole was, of course, increased to give the same total amount per tree.

To shorten the procedure and make it more practicable commercially, small pellets of manganese sulphate were used in 1941. Further, their use prevented contact of the salt, previously used in powder form, with the bark, thus ensuring quicker healing of the hole.

Satisfactory results were obtained only when the full doses and the full number of holes recommended by Bennett (one for each inch of the diameter of the trunk) were employed. Where reduction of the number of holes was attempted, no tree benefited similarly in all its branches and some received no benefit from the injection. No injury to the leaves occurred through excess of the salt and in all cases healing of the holes was good. Some holes were healed by August, 1941, and by October several more were healed, but the majority did not heal until the spring of 1942.

6. *Solid injection of whole trees in* 1942.

In 1942 a further attempt was made to simplify and shorten the procedure of solid $MnSO_4$ injection. Tablets of dry manganese sulphate, each just less than half an inch in diameter, $\frac{1}{4}$ in. long, and weighing one gramme (instead of pellets weighing only one-eighth of the amount used in 1941) were prepared without a binding matrix. They were inserted into holes drilled with a half-inch Irwin bit, held in an ordinary wooden brace, spaced around the trunk at intervals, one hole for each 1 in. of the diameter of the trunk, at any convenient height. After insertion of the prescribed dose the holes were closed with thin corks as before.

In April, 1942, about 80 mature trees were thus treated, and in May and June of that year all of them bore dark green leaves. New shoot growths, from four to eight inches long, were produced where previously less than one-inch growth had been made during several years and many of the trees carried moderate crops of fruit, much greater than those in many previous years. It remains to be seen, of course, for what period of time the very definite amelioration in the health and cropping of the trees will persist ; but judging from the results obtained with branches injected in March, 1939, at least a four-year period may confidently be expected.

ACKNOWLEDGMENTS.

The writer is greatly indebted to Dr. W. A. Roach, Mr. S. G. Thompson, Mr. B. S. Furneaux, Dr. N. H. Pizer, Mr. W. G. Kent and Mr. C. R. Thompson for their valuable help throughout the experiments and in the writing of the paper.

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SUMMARY.

Certain decrepit cherry trees, in well-defined areas of an orchard in Kent, showed, amongst other signs of unhealthiness, marked chlorosis. The variety most badly affected was Governor Wood; Amber Heart and Roundel Heart were less so; Early Rivers and Biggareau Napoleon least.

Spectrographic analysis of samples of the leaves suggested serious manganese deficiency and possibly slight deficiency of magnesium.

The affected trees grew on a shallow, eroded, soil with bad drainage, the others in the same orchard on well-drained Valley Brickearth. The pH value was the same for both soils. Chemical analysis showed less than half as much manganese in the soil under the chlorotic trees as under healthy trees. Injections through leaf-stalks and shoot-tips of solutions of salts of Mn, Mg and Fe confirmed the spectrographic diagnosis; the case was not one of iron deficiency.

An attempt to cure the trouble by generous applications of farmyard manure failed.

Spraying the affected trees with solutions containing manganese sulphate caused slight improvement but insufficient for practical purposes.

Injecting branches and whole trees with solutions of manganese sulphate gave slightly better results, but, for some unknown reason the quantity of solution absorbed was too small to be really effective.

Injection of large branches and tree trunks with solid manganese sulphate resulted in full restoration of the green colour to the leaves and marked increase in growth and in the crop of fruit. It is not yet known whether these satisfactory results will be permanent, but it is confidently expected that they will last for a period of four years at least.

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PLATE I.

a b c d e

FIG. 1.

Leaf series showing from left to right progressive symptoms of manganese deficiency in cherry leaves.

FIG. 2.

Top Branch: Injected with solid manganese sulphate in March, 1939, and photographed the second season after injection.

Bottom: Control from same tree, not injected.

PLATE I.

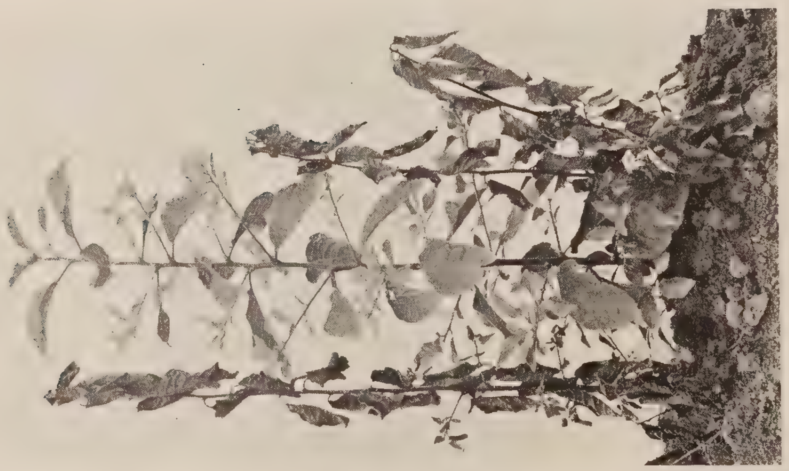

FIG. 1.

Part of plum layer row, showing three withered shoots and one healthy one.

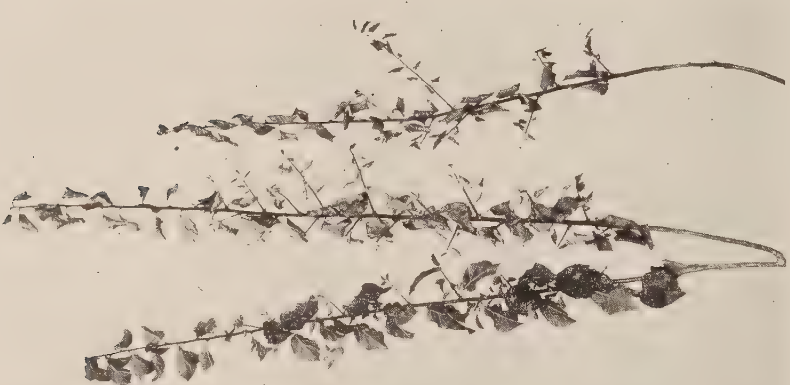

FIG. 2.

Plum shoots pulled up from an infected layer row. From left to right: healthy shoot, one beginning to wilt, one with leaves withered.

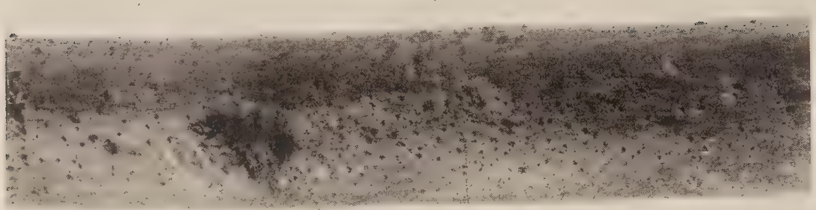

FIG. 3.

A lesion as seen with a pocket lens, showing microsclerotia. $\times 4\frac{1}{2}$

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FIELD OBSERVATIONS ON THE CYLINDROCLADIUM SHOOT WILT OF PLUM AND CHERRY LAYERS

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WILTING of shoots of plum layers and stools in recent years has been so severe on some of the most valuable rootstock varieties propagated at the East Malling Research Station that measures for controlling it are now an urgent necessity, particularly since these varieties are being propagated in quantity for post-war distribution. As the loss from this disease may in some seasons attain to 40 or 50 per cent. of the shoots the seriousness of the problem is evident.

A wilting of plum layer shoots was first noticed at the East Malling Research Station in 1924, and the disease at that time was shown to be due to infection of the leaves, buried during earthing up, by *Monilia cinerea* (Wormald, 1935). After two or three years this disease was no longer seen on that part of the Station, but in 1932 a similar type of wilting was observed in another nursery bed and later in other fields on the Station. On the earthed-up parts of the shoots lesions were found bearing microsclerotia of a fungus, which, on subsequent isolation in pure culture, proved to be quite distinct from *Monilia cinerea*, and was later (Wormald and Harris, 1937) found to be a species of Cylindrocladium. In inoculation experiments with pure cultures of this fungus wilting was caused comparable with that seen in the field.

SYMPTOMS.

In some years the wilting is first observed as early as the middle of June, but it is usually most noticeable during July and early August. Later it is less conspicuous, as further infection ceases and the wilted shoots become overtopped, and so almost, or completely, hidden by the healthy shoots. During July, shoots at various stages of wilting are to be seen, and on the plum varieties may be noticed shoots with (1) the lower leaves curled upwards at their edges and showing yellow blotches, (2) the lower leaves yellow, the upper ones curled but green, (3) the lower leaves yellow and flagging, the upper ones green and not flagging, (4) all the leaves flagging, (5) all the leaves completely withered. (Plate I, Figs. 1 and 2.)

Yellowing of the foliage is usually the first symptom, but in the Mahaleb cherry, which suffers from the same disease, the leaves of affected shoots become creamy white, rather than yellow, before they begin to wilt.

These observations, together with the results of inoculation experiments, indicate that the wilting in the field occurs chiefly as a result of the heavy earthing-up carried out at the end of May or the beginning of June, though the second heavy earthing-up (early July) probably accounts for some of the later wilting. That the disease is directly correlated with the earthing up operations was shown in 1939 on a plot (about 400 ft. long) of eight layer rows of Common Mussel plum, where one and a half rows were left uncovered during the summer, while the other half row and the rest of the whole rows were earthed up in the usual way. There was no wilting whatever on the one and a half rows not earthed up ; in the half row earthed up there were thirty wilted shoots, and in the other whole rows the number of wilted shoots varied from 60 to 130, out of a total of approximately 2,000 per row.

Until 1940 the disease was not serious in the commercial layer beds of plums on the Station, but in that year it was very destructive to two varieties—Brompton, about 40 per cent. shoots killed, and St. Julien A, over 50 per cent. Previously, in 1935 and 1936, it had been very severe

on an experimental plot containing a number of varieties of plum, cherry, peach, and apricot. Of these it was worst on the following varieties of plum and cherry, the percentage of dead shoots being given in brackets :

Plums. Kroojespruim (22), Black Damas A, (31), White Damas (34), Narrow Leaved Shiny Mussel (36), Myrobolan, selection from California (51), St. Julien J (57), and Bastard Common Mussel (62).

Cherries. Two selections from seedlings of wild cherry (21 and 49), three selected varieties of Mahaleb (27, 32 and 43), and Kentish cherry stock from New Zealand (41).

Some of the above are not propagated on a commercial scale, but among the varieties largely grown for rootstocks, Brompton, St. Julien A, and Myrobolan B have proved to be very susceptible, and the disease occurs also on Common Plum, Common Mussel (A, B and C), Broad Leaved Shiny Mussel, Myrobolan (A, C, D and E), Black Damas (A, C and D), and St. Julien (E and J). It will be noted that in a severe attack 50 per cent. or more of the crop of rootstock shoots may be destroyed.

Rootstocks of quince and apple have not shown this wilting although they have been layered in the same fields as those of plum and cherry.

If wilting shoots are pulled they generally come up whole by separating at the base from the layer, but sometimes they break off at a lesion. The leaves of the parts below ground have by this time almost or entirely decayed and disappeared from both healthy and diseased shoots. The lowest 3 or 4 cm. of the wilted shoots are usually alive, and the lesions themselves are from 2·5 to 9·5 cm. long. The infected tissues of shoots with flagging leaves are dark brown, but on shoots that have been dead for some time the lesions are mostly silvery grey, and the microsclerotia show up clearly, when examined with a lens, on the pale grey areas. Wilted shoots are always completely girdled by the lesions. On one occasion a healthy looking shoot, accidentally pulled up with a wilted one, bore a lesion which had only half girdled it, and callus was forming around the edge, showing that further extension of infection in the shoot had ceased. Apparently, therefore, infection does not always extend right round a shoot and kill it ; such half-girdled shoots would be unnoticed, since their parts above ground develop more or less normally and show no wilting.

The fungus.

The fungus readily produces microsclerotia on the lesions (Plate I, Fig. 3) and also in agar cultures. Its fructifications, however, have not yet been seen on artificial culture media, though some have been observed on a few naturally infected layer shoots of apricot, plum and cherry, and more were found on plum cuttings in a frame. These fructifications are branched conidiophores each bearing a cluster of cylindrical, colourless, two-celled conidia, measuring about $50 \times 5\ \mu$. On many conidiophores the central axis is prolonged beyond the spore-cluster and terminates in a sterile knob. The spores, when sown in culture media, produce growth similar to that obtained from isolations made direct from tissues of the lesions.

Until 1940 the disease had been found in Britain only at East Malling, but in that year a similar Cylindrocladium was isolated from wilted shoots of a cherry rootstock received from Wisley, Surrey.

A more detailed description of the fungus and an account of successful inoculation experiments will be given in a later paper, but it may be mentioned here that in the morphology of its fructifications the parasite conforms to *Cylindrocladium scoparium*, first found by Morgan (1892) on a pod of Gleditschia, and later described by Massey (1917) as the cause of Crown Canker of roses in the United States. In pure cultures, however, the shoot wilt fungus has a habit very different from that described by Massey from roses ; so, until a more detailed comparison

has been made, it may, for convenience, be considered to be a variety of *Cylindrocladium scoparium* Morgan that can be distinguished by cultural methods.

CONTROL MEASURES.

The fungus has not been seen on the aerial parts of the plants in the field; hence it is assumed that infection comes from the soil and that the parasite is a soil organism.

So far as is known the disease attacks the young shoots only, so that if these survive their first year they can be planted out for rootstocks without fear that they will become infected later. Control measures, therefore, will have the object of (1) preventing the transmission of the disease to new sites when clonal stocks are transplanted for raising new plots of layers or stools, and (2) eliminating, or reducing the intensity of, the disease in layer beds where it has already become destructive. The study of the disease and its cause suggests certain precautions that should be taken to prevent it from spreading to fresh layer beds. Where it has become established, control will involve either soil treatment or the application of a fungicide to the growing shoots before earthing-up to protect them from infection.

When the disease is present in a particular field it will be unwise to transplant material for layering direct from that field to another, since there is the risk that the soil clinging to the roots will contain the mycelium, spores, or sclerotia of the fungus, which will thus be transferred to the new site. It is suspected that, in the past, new outbreaks have arisen by transplanting from old affected layer plots to new ones. Such transmission of the disease may be obviated in two possible ways, though neither has yet been tested. If rootstock material must be taken from a field where the fungus is known to be present, the lower ends of the shoots should be dipped in a fungicide (e.g. Bordeaux mixture) before they are planted up. If there is no urgency for planting up a large area immediately, it is suggested that cuttings from aerial parts of shoots should be planted in sterilized soil, and when these have rooted and produced shoots of sufficient vigour they could be planted out on a new site to form the nucleus of a fresh set of layers for further propagation.

Some of these varieties of plums (e.g. Brompton and St. Julien A), however, are propagated only with difficulty from cuttings, and, as the demand for plum rootstocks may soon be on a considerable scale, propagation by layering must be continued ; hence treating the layer shoots to prevent infection from the soil should be adopted. Bordeaux mixture might be effective, but it has not yet been tried on rooted plum shoots ; it would probably prove harmless to them since a 1 per cent. Bordeaux mixture has been used in Russia with safety when dipping tobacco seedlings for the control of diseases (Grooshevoy and Popova, 1940). Experiments will be carried out to test the value of dipping the shoots in a fungicide. Meanwhile, thoroughly washing the roots in clean, preferably running, water, to free them from soil which may contain the fungus is recommended. It may be that this method in itself will be sufficiently effective, but this can be determined only by trial. Spraying the shoots with Bordeaux mixture immediately before earthing up has given as yet inconclusive results.

SUMMARY.

1. A wilting of shoots in layer rows of plum and cherry varieties raised for rootstocks has been found to be caused by a fungus which, at present, is considered to be a cultural variety of *Cylindrocladium scoparium* Morgan.

2. The fungus fructifications are rarely found on the wilted shoots although microsclerotia usually occur on them.

3. The fungus has been isolated directly from lesions on the wilted shoots and from conidia and proved to be parasitic, but details of the experiments are reserved for a future paper.

4. Certain control measures are tentatively suggested.

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DRY EYE ROT OF APPLES CAUSED BY *BOTRYTIS CINEREA* PERS.

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Introductory and historical.

During the past six years the writer has had under observation a disease of apples to which the name Dry Eye Rot was given in New Zealand in 1940 (1). This name is retained here in order to distinguish the disease from another Eye Rot of apples, caused by *Cylindrocarpon mali* (Allesch.) Wr. (4). Laxton's Superb appears to be the most susceptible variety, but comparatively severe attacks have been observed also on Worcester Pearmain, Bramley's Seedling and John Standish.

A few reports on what appears to be the same disease have appeared in the literature. Thus, Cunningham, in 1925 (3), described a rot of apples in New Zealand caused by *Botrytis cinerea*, the initial infection starting in the eye. Again, in 1940, the Dry Eye Rot referred to above and attributed to *B. cinerea*, was reported in New Zealand (1) as affecting fruit of Jonathan and Cox's Orange Pippin, and this led to the rejection of the attacked fruits for export because of the possibility of the occurrence of further rotting in transport. Finally, Wormald, in 1940 and 1941 (7, 8), described a similar type of rot of apples in England which, he stated, appeared to be the result of infection by *B. cinerea*. In no instance hitherto, however, has it been proved by experiment that this fungus is actually the cause of the disease.

Symptoms and course of the disease.

A large number of apples suffering from Dry Eye Rot have been examined by the writer, and the inception and development of the disease has been found to be similar in all the varieties under observation. In its initial stage it is not unlike the Eye Rot caused by *Cylindrocarpon mali*, but it differs from this in its later stages owing to an outstanding characteristic, namely the arrested development of the lesion, which typically becomes hard and dry, although under certain conditions the attack may proceed and develop into a soft rot involving the whole fruit.

Although descriptions of the static phase of Dry Eye Rot have been given (7, 8) its further course of development has apparently not previously been observed. In the summer of 1941 the initiation and subsequent development of the disease were studied on fruits still on the tree and, later, when in storage. Three stages may conveniently be distinguished:

(1) Towards the end of July a slight red colour appears at the base of one or more of the sepals. The reddened area enlarges slowly radially, the skin over it turning light brown; and this increase in area may continue until the first week in September. By this time the lesions vary in diameter from ½-1 in. Internally, the tissues beneath the discoloured skin are soft, water-soaked and light brown in colour to a depth of 1/10th in.

(2) The lesions then cease to enlarge and gradually dry out. The skin overlying them slowly subsides, turns dark brown or black and assumes a stretched parchment-like appearance, frequently becoming detached from the surrounding healthy skin. Internally, the previously water-soaked tissues also dry out and become tough. The lesions at this stage are approximately circular and either surround the calyx, or, more frequently, lie to one side of it. Their margin is clearly defined and is frequently surrounded by a narrow band of red skin. This phase of the disease is shown by the upper two apples in Plate I, Fig. 1.

(3) The lesions remain in the quiescent condition reached at the end of stage (2) for a number of weeks. Subsequently, during October and November, the healthy skin immediately surrounding the lesions becomes light brown; internal decay starts again and spreads until,

after about fourteen days, the whole apple is involved in a light brown, spongy, soft rot, an aromatic odour being emitted. The lower two apples in Plate I, Fig. 1, illustrate this stage.

In nature, while the apples are on the tree, stage (1) is followed by stage (2) although, under exceptionally wet conditions, stage (1) may pass directly into stage (3) without the intervention of the quiescent stage (2), as was observed during September, 1942, in Laxton's Superb, growing in a Worcestershire orchard. Again, stage (3), which usually appears when the apples are in storage, does not necessarily follow stage (2). Apples in stage (2) have usually shown no further development of the disease after being in storage for five months; on the other hand, in one particular sample of 178 infected Laxton's Superb, 123 of the fruits developed total rots (stage (3)) during storage whilst the other 55 remained in stage (2).

Isolation of the fungus.

The static stage of Dry Eye Rot was found frequently in fruit still on the tree during the seasons of 1937-40, and in this period two methods of isolation were used:

1. *From rotted tissues.*—Affected fruits were submerged in 1/1000 $HgCl_2$ for five minutes, washed in running water for ten, rinsed with absolute alcohol and flamed. Small pieces of diseased tissue were then teased out and plated on 2 per cent. malt-extract agar. From 83 attempted isolations, 56 gave pure cultures of *B. cinerea*, 24 remained sterile and 3 produced a Penicillium. Most of the attempted isolations which gave no growths were made from affected apples late in the growing season or during the first month of storage.

2. *From spores.*—Spores on affected sepals and on the surface of the lesions were detached with a sterile needle and placed on 2 per cent. malt-extract agar. The 23 isolations attempted yielded a pure culture of *B. cinerea* in each instance.

In August, 1941, when the actual course of development of Dry Eye Rot was followed, a further nine isolations from the affected tissues in stage (1) (which had been found to be invaded by septate mycelium) also yielded pure cultures of *B. cinerea*. The tissues from the total rot described in stage (3) yielded the same fungus.

Inoculation experiments and results.

Using the fungus isolated from stage (1) of the disease, a series of inoculations was carried out during the summers of 1941 and 1942. On August 24th, 1941, small punctures were made with a sterile needle at the base of the sepals of 47 healthy Laxton's Superb apples while still on the tree. Spores and mycelium from a ten-day-old culture were inserted into and placed over each wound, and each inoculation was covered for ten days by a piece of dry cotton wool, held in position by an elastic band.

In the laboratory, small portions of inoculum were inserted into wounds made in the skin of four varieties of apples which were then placed in moist chambers.

Control apples of both types, wounded but not inoculated, were included in the experiment.

Of the 47 apples inoculated on the trees 45 failed to show any rot. The remaining two showed small lesions after ten days, which expanded gradually until the fruits were completely rotted. They were soft, and isolations from the watersoaked tissues gave pure cultures of *B. cinerea*.

The inoculations on the four varieties in the laboratory produced soft, light brown rots, which eventually involved the whole fruits. Isolations from the rotted tissues again gave pure cultures of *B. cinerea*.

Thus, of 51 apples originally inoculated with *B. cinerea*, only 6 became infected, and stage (2) of Dry Eye Rot was not exhibited by any of them. All the controls remained sound.

The culture of *B. cinerea* isolated during 1941 was kept in an actively growing state on malt-extract agar during the ensuing winter and was used in 1942: (1) To ascertain whether *B. cinerea* is actually the causal organism of Dry Eye Rot. (2) To determine why the rot generally dries out instead of continuing to extend and involve the whole apple. (3) To compare

the attack of the fungus on fruits having intact calyces with that on fruits with mechanically damaged calyces.

Fruits still attached to the trees of Cox's Orange Pippin and Laxton's Superb were selected for this work. For inoculation, 7-8 day-old sub-cultures of the fungus, grown on plates of 2 per cent. malt-extract agar, were always used. Pieces of agar 1 cm. square bearing the actively growing fungus and removed with a sterile needle, were placed with the mycelium-bearing surface in close contact with the calyx of each apple. The inoculum was held in position by a strip of wide adhesive tape so placed that the calyx depression of each apple was sealed. In this way the fungus was permitted to grow in a confined moist atmosphere.

After periods varying from three days to a week the tape and the remains of the inoculum were removed and the apples were marked with white paint and kept under observation. The first inoculations were made on July 10th, 1942, after the close of a comparatively late " June drop ", and the last on August 28th.

The inoculum was placed on the calyces of apples of three types :

(1) Those with uninjured still living sepals.

(2) Those with still living calyces, wounded by the forcible removal of one or more sepals.

(3) Those with uninjured calyces, the sepals of which were dead and had dried up naturally.

Control apples similarly treated but not supplied with the inoculum were also included.

The results of the experiments are given in Table I.

TABLE I.

Summary of inoculation experiments, 1942.

Type of inoculation.	Variety.	Date of inoculation.	Date of removal of seal.	No. of fruits inoculated.	No. of fruits infected.
(1) On uninjured living calcyes	Cox's Orange Pippin	15.7.42	22.7.42	21	0
do.	Laxton's Superb	16.7.42	23.7.42	20	0
(2) On injured living calyces	Cox's Orange Pippin	10.7.42	13.7.42	19	19
do.	do.	do.	15.7.42	34	34
do.	Laxton's Superb	do.	16.7.42	30	30
(3) On uninjured dead calyces	Cox's Orange Pippin	28.8.42	4.9.42	19	0
do.	Laxton's Superb	do.	do.	15	0

It will be seen that the only apples to become infected were those in group (2) in which injury had been caused to still living calyces. On these apples the following observations were made :

Cox's Orange Pippin.—The removal of the tapes showed that circular, medium-brown lesions, $\frac{1}{4}$-$\frac{1}{2}$ in. in diameter, had developed. Those on the apples from which the tapes were removed after five days had an average diameter slightly in excess of those on the fruits from which the tapes were removed two days earlier. After about a fortnight all the lesions had practically dried out, and the overlying skin was somewhat sunken, black and hard. At the margin of most of the lesions, splitting away from the healthy skin had occurred (Plate I, Fig. 2). The depth of penetration of the fungus was no greater than $\frac{1}{8}$th in. No increase in the diameters of the lesions took place after removal of the tapes and no further change in them occurred up to September 17th. Some of the fruits had then fallen owing to wind and Codling attack.

Isolations from infected tissues of the pre-dry-out stage, on July 16th, and from spores on the surface of the dried out stage, on July 30th, gave pure cultures of *B. cinerea*.

Laxton's Superb.—Twenty-seven of the thirty apples showed lesions with diameters of $\frac{1}{2}$-$\frac{3}{4}$ in. and the other three had lesions of $1\frac{1}{4}$ in. in diameter. They were flattened, light brown with a darker brown margin, and internal penetration was little more than 1/10th in. The affected tissues were pale brown and water-soaked. The smaller lesions had completely dried out by July 27th and the overlying skin had become somewhat sunken, black, hard and contracted away slightly from the surrounding healthy skin. The three larger lesions were still expanding and had involved half the surface of the fruits. Later, total rot occurred in these fruits and they fell from the trees.

By August 28th, ten of the other twenty-seven apples had been blown down. That day was extremely hot and the seventeen apples still on the tree were growing rapidly. As a result, comparatively large cracks appeared in them around the lesions, and the exposed flesh was being severely attacked by wasps ; moreover, signs of infection by *Sclerotinia fructigena* were becoming evident. On September 17th all the apples had become completely rotted by this fungus and some of them had fallen. Isolations made from the rotted tissue of the pre-dry-out stage and from spores appearing later on the surface of the dried out lesions, before the secondary attack by *S. fructigena*, yielded pure cultures of *B. cinerea*.

The control apples in both varieties failed to develop lesions.

DISCUSSION.

Out of a total of 115 isolations attempted from the three naturally occurring stages of Dry Eye Rot, *B. cinerea* was obtained in the pure state on 88 occasions. Twenty-four attempted isolations from the rotted tissues of stage (2), most of which were made late in the growing season, remained sterile, a fact which suggests that the fungus may be killed as a result of desiccation ; and this would explain why stage (3) does not always develop when apples showing stage (2) are placed in storage.

As a result of inoculations made in 1941 and 1942 all three stages of the disease were reproduced separately under various conditions although they did not appear in succession. Stage (2), however, was induced to follow stage (1) when 83 apples with injured calyces were inoculated on the tree and kept moist by a tape seal. All the fruits developed stage (1) which, on the removal of the seal after 3-7 days, passed to stage (2). Three of the fruits, however, passed directly from stage (1) to stage (3), as did apples inoculated in the laboratory and kept under moist conditions.

Isolations made from the three stages of naturally occurring and artificially produced Dry Eye Rot, both from infected tissues and surface spores, all yielded pure cultures of *B. cinerea*, a result which justifies the conclusion that this fungus is the causal organism of this disease.

Two parallel cases in which attack by *B. cinerea* remains limited to localized areas of infection are provided by the Chocolate Spot disease of broad and field beans and the Grey Mould of tomatoes. As to the former, Wilson (6) states that two types of attack exist, (*a*) the " non-aggressive " form, causing the typical Chocolate Spot symptoms, and (*b*) the " aggressive " form, which causes blackening and death of part or whole of the shoot system. He found that the " aggressive " form could be induced only under optimum conditions of humidity, rain, temperature and wind, and that in the absence of one or other of them the " non-aggressive " form only would develop. With Grey Mould of tomatoes, Read (5) observed lesions developing " occasionally at the blossom end of the fruit if the large drop of water which sometimes collected there did not evaporate within 24 hours or so, but the rot formed in this manner frequently failed to spread through the entire fruit and the affected portions dried up ".

Similar behaviour occurs in the Fire disease of tulips, caused by *Botrytis Tulipae* (Lib.) Lind (2). The lesions here enlarge rapidly in moist weather but dry out and often split in dry weather. In addition, numerous small spots appear during showery weather on the leaves and

petals as the result of infection by individual spores splashed by rain from Fire lesions. If the fungus remained alive, each spot would be expected to develop into a larger lesion under moist conditions. Attempted isolations from the small spots, however, showed the fungus to be dead in almost every instance, hence the spots did not enlarge and formed a static phase of the disease.

In Dry Eye Rot of apples it is suggested that meteorological conditions may be solely responsible for the cessation in the development of the lesion, as opposed to the suggestion made by Wormald (7) that a protective barrier of cork cells may be formed isolating the infected region ; for the presence of such a barrier could not be detected by the writer after a number of affected apples had been examined in sections. In the present work, stage (1) developed when 83 apples with damaged sepals were inoculated and kept under moist conditions. On the removal of the seals, causing a marked reduction in humidity, 80 of the lesions immediately dried out and developed stage (2), a result which indicates that changes in humidity play an important part in the development and drying out of the lesions.

Inoculations carried out in 1942 clearly indicated that mechanical injury to the sepals must occur before Dry Eye Rot can develop. Wormald (7, 8) suggested that desiccated or dead sepals, when moistened by rain, would easily become attacked by *B. cinerea*, which would then grow into the flesh itself. This view is not supported by the present work, for when thirty-four apples with uninjured, dead calyces were inoculated, all failed to develop any form of rot.

SUMMARY.

A description is given of an apple disease known as Dry Eye Rot. For convenience its course of development has been divided into three stages, viz. (1) the formation of the initial eye rot, (2) the quiescent Dry Eye Rot, and (3) the development of the disease from stage (2) into one involving complete rot of the whole fruit.

Isolation and inoculation experiments are described which establish the fact that *Botrytis cinerea* Pers. is the causal organism of this disease.

Injury to the calyx is necessary before infection can occur and the disease become apparent.

It is suggested that a change from high to low humidity frequently arrests the progress of the disease at the Dry Eye Rot stage and prevents its further development into a complete soft rot of the fruit.

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(*Received* 17/12/42).

PLATE I.

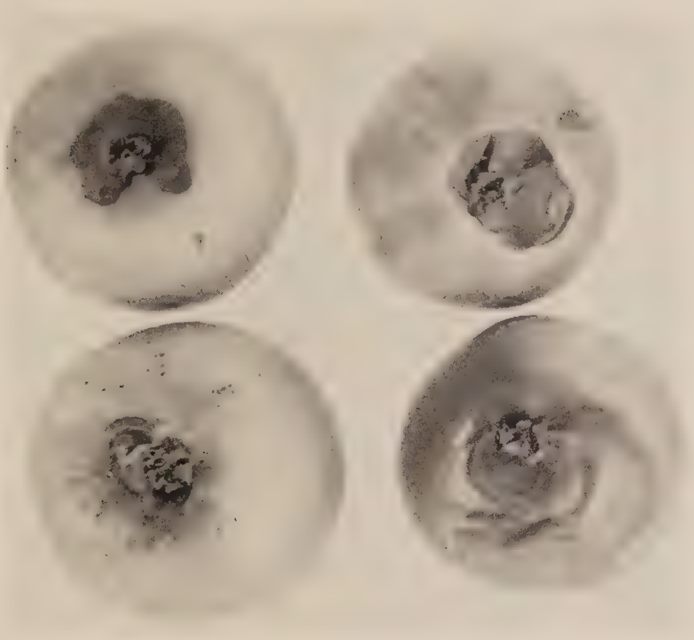

Fig. 1.
Dry Eye Rot of apples in Laxton's Superb.
Upper two apples showing stage (2).
Lower two apples showing stage (3).

Fig. 2.
Dry Eye Rot of apples induced in Cox's Orange Pippin by inoculating apples, having damaged calyces, with *Botrytis cinerea* isolated from stage (1) of the disease.

THE INFLUENCE OF LIME AND POTASH ON MOSAIC INFECTION IN THE TOMATO (var. *POTENTATE*) UNDER GLASS

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INTRODUCTION.

In work reported earlier (Selman, 1941*a* and 1942), experiments to investigate the deleterious effects of tomato Mosaic virus on the tomato crop under glass were undertaken. With some knowledge of the effects associated with this disease, experiments were directed to efforts to mitigate these effects by attention to soil and cultural conditions. The reduction in the numbers of fruit set and the increase in the amounts of fruit showing blotchy ripening were discussed in relation to cultural control measures (1942), and it was considered that one factor which might profitably be studied in seeking to overcome these effects was the balance of fertilizers in the soil.

The effect of the lime/potash ratio on plant growth has been studied by many workers, and it has been found that the type of growth may be markedly affected, apparently through changes induced in the water relations of the tissues (Lundegårdh, 1935 ; Tiedjens and Schermerhorn, 1936). In view of the changes in the water relations of the tomato plant induced by Mosaic infection and also of the large dressings of lime and sulphate of potash that are applied in commercial tomato culture in this country, an experiment was designed to investigate the interaction of Mosaic infection with lime at two, and sulphate of potash at three levels of application.

METHODS.

The same glasshouse was used as in previous work, but to facilitate control of the manurial dressings the plants were grown in 10-inch pots.

Soil.

In the autumn of 1941 the top 5 in. of soil was removed from the borders in the house to eliminate any effects which might accrue from any undecomposed plant residues in the surface soil. The rest of the soil in the borders was steam sterilized in December, 1941, and samples for analysis were taken with a trowel to a depth of 6-8 in. in January, 1942, before the application of base fertilizers.

To eliminate the occurrence of gross variations in its composition, 192 ten-inch pots were filled with the soil from the borders, drawn at random from all parts of the house and thoroughly mixed on a bench before adding the manures.

Manurial treatments.—Each pot contained approximately 12½ lb. of moist soil (equivalent to 9½ lb. of air-dry soil) and to this was added 1 oz. dried blood and 1 oz. superphosphate, together with lime and potash at one of the following rates :

No additional lime	Ca_0
2 oz. slaked lime per pot	Ca_2
No additional potash	K_0
1 oz. sulphate of potash per pot	K_1
3 oz. sulphate of potash per pot	K_3

Top dressings.—The only top dressings applied were dried blood, superphosphate and sulphate of potash, and these were always supplied in the ratio in which they had been included in the

base, but at varying rates. Six dressings were applied, the first on May 4th and the last on August 7th. The total top dressings were:

Dried blood	$1\frac{1}{4}$ oz. per pot to all pots	
Superphosphate	$1\frac{1}{4}$ oz. per pot to all pots	
Sulphate of potash	0 per pot ..	K_0
,, ,, ...	$1\frac{1}{4}$ oz. per pot ..	K_1
,, ,, ...	$3\frac{3}{4}$ oz. per pot ..	K_3

On September 3rd, when the fruit from five trusses had matured, the soil was sampled to the depth of the pot, taking one pot at random from each plot. The analysis of the soil before the experiment and from treatments Ca_0K_0 and Ca_2K_0 at the end of the season is given in Table I.

TABLE I.

Soil analysis calculated for soil dried at 100° *C.*

	January, 1942.	September, 1942.	
	No additional fertilizers.	Ca_0K_0.	Ca_2K_0.
pH	7·72	7·55	7·79
	%	%	%
Total carbonates, as $CaCO_3$	4·39	3·89	4·35
Total K_2O	0·51	0·49	0·52
Available K_2O	0·11	0·03	0·02
Total P_2O_5	1·14	0·80	0·80
Available P_2O_5	0·16	0·31	0·26
Total N_2	0·35	0·40	0·44
Loss on Ignition	12·85	11·30	11·42

No bulky organic materials were included in the soil used for the main experiment, so that its texture was unlikely to have been ideal for plant growth. For comparative purposes, twelve other pots were filled with a standard potting mixture as follows:

1 part decaying stable manure :	5 parts maiden clay loam	
Base fertilizers per pot:	Hoof and horn ..	1 oz.
	Superphosphate ..	1 oz.
	Sulphate of potash ..	1 oz.
	Slaked lime	$\frac{1}{2}$ oz.
Top dressings:	As for K_1 series.	

This treatment approximated rather more closely to nursery practice in pot work and, although not replicated in the main experiment, provided a useful qualitative comparison with the other treatments.

Plant material.

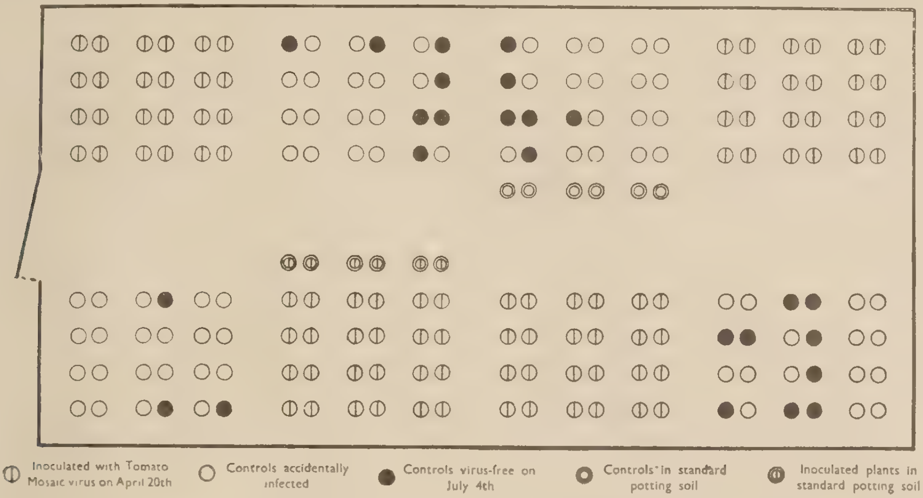
Tomato seed of the variety "Potentate", saved from leaf-tested, virus-free plants, was sown in boxes on January 29th, 1942, the seedlings potted into 3-inch pots on March 5th and transferred to the 10-inch pots on April 9th, when at the ten-leaf stage. It was noted that where lime or sulphate of potash had been included in the soil there was some check to growth at the time of planting out, whereas no check occurred where lime and potash had been omitted.

Design of experiment.

Text Fig. 1 is a diagram showing the arrangement of the pot plants in the house. The house was divided into four blocks, each of which was subdivided, one-half of the plants were to be inoculated with tomato Mosaic virus (A.17) and the other to be uninoculated controls. On April 20th, when the first truss was in bloom, one-half of the plants were inoculated with

this virus by wiping one leaflet of every leaf on the plant with muslin soaked in infective juice taken from Mosaic diseased tomato plants.

Each half of the block consisted of six plots for the manurial treatments, and each plot consisted of four plants. The arrangement of the half blocks within blocks, and of the six plots within the half blocks, was at random.



TEXT FIG. 1.

Diagram to show positions of healthy and infected plants (not to scale).

Spacing.—There were four rows of plants in each border, and the plots were alternately 2 ft. and 3 ft. apart, with a distance of 4 ft. between the half blocks. It was hoped that this arrangement would help to prevent accidental spread of infection to the control plants during cultural operations.

Meteorological data.

The season was above the average for hours of bright sunlight, the number recorded at this Station in 1942 from April-August inclusive, being 994·5 as compared with a ten-year mean (1931-40) for these months of 876·7 hours. This total was fairly evenly distributed throughout the growing period, the daily averages for each month being shown in Table II.

TABLE II.

Daily averages of bright sunlight.

Month.	Hours bright sun per day.
April	7·40
May	6·82
June	7·93
July	5·65
August	4·77

The air temperature of the house was usually maintained between 65-70° F. in daylight, rising above this only with sun heat. The maximum temperature recorded was 94° F., ventilation usually being applied when the temperature exceeded 75° F. Night temperatures were rarely allowed to fall below 58° F., the minimum recorded during the season being 51° F.

The condition of the plants was the indicator used to determine amount and frequency of watering and overhead damping.

DISEASE SYMPTOMS.

Mosaic.—Observations were made of the appearance of Mosaic symptoms on the inoculated plants, and a summary of the numbers of plants showing the earlier symptoms is given in Table III.

TABLE III.

Numbers of plants showing early symptoms of the disease.

Treatment.	Ca_0K_0.				Ca_0K_1.				Ca_0K_3.			
Symptoms.	N.S.	V.	G.M.	Y.M.	N.S.	V.	G.M.	Y.M.	N.S.	V.	G.M.	Y.M.
April 30	0	16	0	0	1	15	0	0	9	7	0	0
May 2	0	11	5	0	0	9	7	0	0	14	2	0
,, 5	0	0	8	8	0	4	8	4	0	8	7	1
,, 8	0	0	0	16	0	0	12	4	0	1	15	0
,, 14	0	0	16	0	0	0	16	0	0	4	12	0
Treatment.	Ca_2K_0.				Ca_2K_1.				Ca_2K_3.			
April 30	6	10	0	0	11	5	0	0	11	5	0	0
May 2	4	12	0	0	6	10	0	0	1	15	0	0
,, 5	0	12	4	0	1	6	9	0	2	11	3	0
,, 8	0	0	16	0	0	1	15	0	0	4	12	0
,, 14	0	0	16	0	0	0	16	0	0	4	12	0

N.S. = No symptoms of virus infection.
V. = Hardening, with slight crimping of the apical leaves. No foliar mottle.
G.M. = Light and dark-green foliar mottle appearing.
Y.M. = Severe yellow-green foliar mottle appearing.

It should be noted that the first top dressing was applied to all plants on May 4th, which may explain the fairly uniform, green foliar mottle noted ten days later.

It will be seen from Table III that symptom appearance was delayed where lime had been applied. The incubation period was shortest in the plants that made the most rapid growth, viz. those of the Ca_0K_0 group, and was longest where lime and potash applications had retarded growth. The type of growth made by plants receiving these manurial treatments is illustrated in Figs. 1 and 2, Plate I. A severe yellow-green mottling was noted for a short period on the more vigorous plants receiving no additional lime, but on no plant was this found where lime had been applied.

Other symptoms.—Later in the season there were indications of the effect of the manurial treatments in a few plants of certain groups. Thus, on July 14th, the following symptoms were noted :

(1) Yellow, glossy patches on the half-grown leaves of inoculated plants receiving lime and high potash (Ca_2K_3).

(2) Orange patches (not glossy) on the older leaves of control plants receiving lime but no potash (Ca_2K_0).

(3) Lemon-yellowing of all leaves. In young leaves, yellowing began at the margins, and pale-brown necrotic spots developed ; old leaves showed purple interveinal and marginal areas. These symptoms were confined to plants receiving neither lime nor potash and undoubtedly indicated a true potash deficiency.* Only three or four plants were thus affected and no relation between these symptoms and Mosaic infection could be discerned.

* Leaf symptoms of potash deficiency were thus evident when the available K_2O (soluble in 1% citric acid) approached 0·03–0·02% (see Table I).

(4) In general aspect the chief difference noticeable between the control and the inoculated plants was the somewhat poorer leaf development of the latter. The leaf span was less, and the area of individual leaflets tended to be smaller in the inoculated plants. Growth in height was not appreciably checked by Mosaic infection. The appearance of the plants on July 23rd is shown in Figs. 3 and 4, Plates I and II. At this stage the plants had been stopped, the fifth truss had set fruit, and ripe fruit was being picked from the third and fourth trusses.

SPREAD OF DISEASE TO THE CONTROL PLANTS.

Earlier comparisons made between inoculated and uninoculated plants have not given the true relation between healthy and Mosaic infected plants, and an attempt was made in this work to study the degree of infection of the control plants when the fifth truss had set fruit. It had not previously been found possible to maintain the controls free from Mosaic infection beyond the stage at which the fifth truss was blooming, despite rigorous precautions. In the present work the control plants were always handled first, and the hands of the worker were washed with soap and water between each half block. As stated above, a distance of 4 ft. was allowed between control and inoculated plants. Removal of top soil and steam sterilization of the soil used in the pots might have been expected to have removed sources of infection, if any, in the soil. The plants had been raised from virus-free seed saved by the author from plants shown to be free from tomato Mosaic virus by wiping juice from their crushed leaves on test plants of *Nicotiana glutinosa* and seedling tomatoes. The plants were handled by non-smokers. Thus, reasonable precautions had apparently been taken to prevent accidental contamination of the control plants ; nevertheless, one such plant showed symptoms of Mosaic infection on June 7th, i.e. seven weeks after the introduction of virus into the house via the inoculated plants. It is uncertain whether insects are able to transmit this virus. A light infestation of red spider mite (*Tetranychus telarius* L.) and a few tomato moths were noted, however.

On July 4th, when Mosaic was widespread on many of the controls, all the uninoculated plants were tested for virus infection by removing one leaflet from the top of the plant and another from a mature leaf near its base. The two leaflets were crushed together in a little distilled water and the juice was wiped on test plants of *N. glutinosa* and on tomato seedlings. The results of these tests are shown in Table IV.

TABLE IV.

Tests for presence of Mosaic virus in uninoculated plants.

Treatment.	No. of plants, out of 16, virus-free on July 4th.
Ca_0K_0	8
Ca_0K_1	2
Ca_0K_3	3
Ca_2K_0	10
Ca_2K_1	2
Ca_2K_3	0

Statistical examination of the data from the individual plots indicated the high significance of the potash effect ("z" = 1·2596, 0·1% point = 1·2141). No significant effect could be attributed to lime level, or to the interaction between lime and potash. Thus, where no sulphate of potash had been applied, the control plants tended to remain immune from accidental infection with Mosaic. The distribution of the healthy plants on this date is indicated in Fig. 1 by the black circles.

FLOWER AND FRUIT PRODUCTION.

Counts were made of flower buds, fruits and chats, and records taken of fruit weights and quality, including unblemished ripe fruit, blotchy fruit, fruit with greenback and fruit showing

blossom end rot. These data were recorded separately for each truss on the plant up to five trusses, the plants being stopped one leaf above the fifth truss.

Statistical analysis of data.

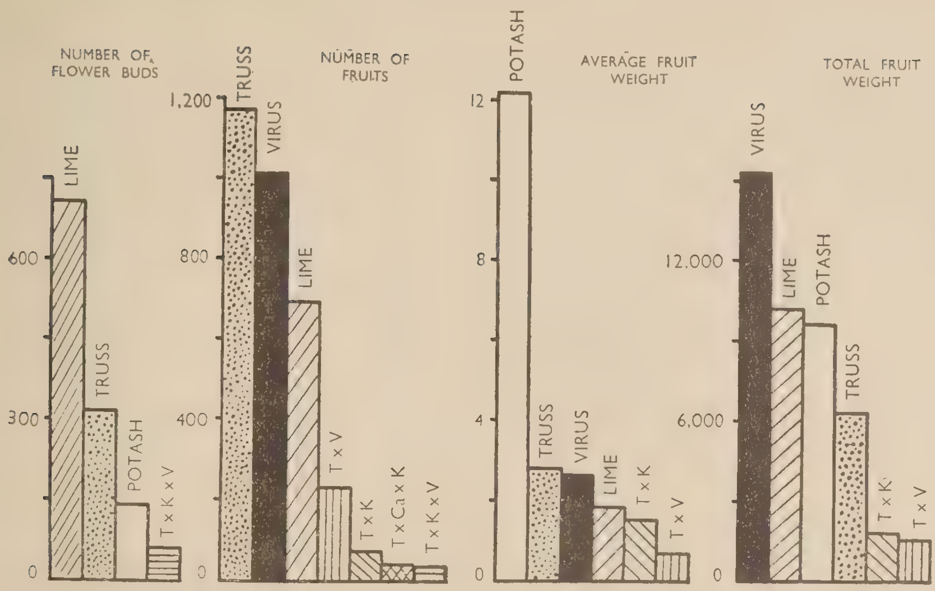
As five trusses were measured separately, the truss, and not the whole plant, was used as the unit for analysis; thus, the variance shown for factors Between Plots represents one-fifth of that obtained for the whole-plant plot totals. One example of the method of analysis is given in detail for Total Weight of Fruit.

Analysis of Variance. Total weight of Fruit.

	D.F.	Mean Square.	"z."
Between Plots.			
Blocks	3	276·67	
Inoculated *v.* Control	1	15,312·04	**1·8587** (0·1% pt. = 1·2936)
Ca_0 *v.* Ca_2	1	10.205·12	**1·6560** (,, ,, ,,)
K_0 *v.* K_1 *v.* K_3	2	9,582·07	**1·6243** (0·1% pt. = 1·0859)
Interactions:			
Virus × Lime	1	23·42	
Virus × Potash	2	127·95	
Lime × Potash	2	287·81	
Virus × Lime × Potash ..	2	87·36	
Deviations (*a*)	33	372·05	
Within Plots:			
Trusses	4	6,256·69	**1·7623** (0·1% pt. = 0·7648)
Interactions:			
Trusses × Virus	4	1,553·56	**1·0657** (,, ,, ,,)
Trusses × Potash	8	1,792·66	**1·1374** (0·1% pt. = 0·5917)
Trusses × Lime	4	286·14	
T × Ca × K	8	246·80	
T × Ca × V	4	55·57	
T × K × V	8	254·38	0·1609 (5% pt. = 0·3309)
T × K × V × Ca	8	129·85	
Deviations (*b*)	144	184·44	
Total	239		

The mean values obtained for the five-truss totals of flower buds together with the number, weight and quality of fruit are presented in Table V. For comparison, the mean values obtained from the plants grown in the standard potting soil are also shown, but these values have not been considered in the statistical treatment of the data, since there was no replication within this group. Where the mean values obtained for the separate trusses have been deemed of interest, the results have usually been expressed graphically. The complete primary data of the experiment have been deposited in the Natural History Museum, South Kensington, London, S.W.7, for consultation by those interested.

An outline of the results of the statistical examination may conveniently be summarized graphically by representing the magnitude of all mean square values for treatment differences which have been found to exceed the 5 per cent. point of significance for "z", by the height of columns plotted side by side for each character measured. This has been done in Text Fig. 2 and illustrates the relative contribution of treatment differences to the total variance of the character measured. Thus, for example, one may say that in this experiment, of the treatments studied, the level of potash is the one predominantly influencing average fruit weight, whereas the position of the truss, the level of lime or the health or Mosaic infection of the plant affected this character to a much less extent. As in the analysis of variance given above, the mean squares due to V, K and Ca (i.e. the treatments effective between plots) are reduced to one-fifth of the actual plot variance to make them comparable with the values for truss position and truss interactions.



TEXT FIG. 2.

Diagram to show relative variance of significance treatment differences.

TABLE V.

Flower and Fruit Production. Totals for five trusses. (Mean values for 16 plants.)

Manurial treatment.		No. of flower buds.	Unblemished ripe fruit.		Blotchy fruit.		Greenback fruit.		Fruit with blossom end rot.		Fruit unripe on Sept. 2nd.		Fruit total per plant.	
			No.	lb. oz.	No.	lb. oz.	No.	lb. oz.	No.	lb. oz.	No.	lb. oz.	No.	lb. oz.
Ca_0K_0 ..	C.	42·6	18·8	4 8½	10·5	1 15	0·8	– 3	0·1	– ½	3·1	– 5½	33·3	7 0
	I.	38·9	13·1	2 10½	11·0	2 5	1·1	– 4	0	0 0	1·5	– 2½	26·7	5 6
Ca_0K_1 ..	C.	37·4	25·3	5 3	3·0	– 9	0·7	– 2	0·3	– ½	2·6	– 6½	31·9	6 5
	I.	38·1	18·1	3 10	5·3	– 15½	1·0	– 4½	1·3	– 2½	2·9	– 4½	28·6	5 5
Ca_0K_3 ..	C.	36·7	17·8	3 4½	3·0	– 9	1·2	– 4	6·4	– 8	2·1	– 4½	30·5	4 14*
	I.	36·6	19·1	2 11½	2·2	– 6	0·7	– 2½	3·2	– 4½	2·2	– 4½	27·4	3 13
Ca_2K_0 ..	C.	35·1	16·8	3 10	6·8	1 7½	1·4	– 5½	0·3	– 1	2·9	– 5½	28·2	5 13½
	I.	34·4	14·9	2 13	6·6	1 6½	0·4	– 1½	0·1	– 0½	1·7	– 2½	23·7	4 8*
Ca_2K_1 ..	C.	35·5	21·8	3 14½	2·3	– 7	1·0	– 2½	1·5	– 3½	3·4	– 6½	30·0	5 2
	I.	34·9	15·7	2 13	3·1	– 8	1·1	– 3	0·6	– 1½	2·5	– 3½	23·0	3 13
Ca_2K_3 ..	C.	33·8	18·3	3 1	2·9	– 8	0·9	– 3½	2·8	– 4	2·4	– 3½	27·3	4 4
	I.	30·9	15·2	2 4½	1·7	– 4	0·8	– 3	1·7	– 2½	1·7	– 2½	21·1	3 0½
Standard potting mixture	C.	39·3	26·8	4 14	1·3	– 3½	1·3	– 3½	0·3	– 1	3·0	– 6½	32·7	5 12½†
	I.	38·0	19·4	3 10½	3·3	– 10½	0·5	– 1½	0·5	– 1½	1·5	– 3	25·2	4 11†

C. = Uninoculated controls.

I. = Inoculated with tomato Mosaic virus when first truss in bloom.

* Mean of 15 plants.

† Mean of 6 plants.

The influence of Mosaic infection on the five-truss totals.

Text Fig. 2 is indicative of the important contribution which health or Mosaic infection of the plant made in this experiment to the final yield of fruit. The number of flower buds was, however, unaffected by the virus, the primary effect apparently being the reduction in the total numbers of fruits, together with a slight reduction in average fruit weight. This confirms the results previously reported with this virus (Selman, 1942).

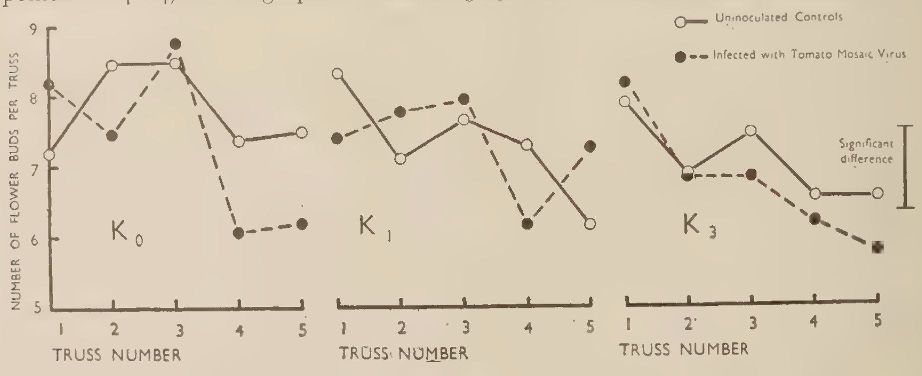
The reduction in the total yield of fruit occasioned by early inoculation with the virus was 22·5 per cent. (Mean of all treatments) as compared with the uninoculated controls. The absence of any significant interaction between Mosaic infection and the level of lime or potash for the whole plant totals, indicated that the effect of the virus on yield reduction was approximately the same at all levels of lime and potash.

The value attributed by growers to cultural control measures is seen from this experiment to be well founded, for well-grown plants infected with the virus when the first truss was in bloom produced over 5 lb. of fruit (Table V), and this would be accepted by the grower as a profitable crop. Plants indifferently manured, but which did not become infected until much later in the season, produced less fruit than these early-infected ones. Other factors being equal, however, there is abundant evidence to show that it is more profitable to grow a virus-free crop.

The interaction between truss position, potash level and Mosaic infection.

When the position of the truss is taken into account, there is some evidence that the effect of virus infection may be determined by the level of potash manuring, particularly in relation to flowering and fruit set. Significant interactions were found as follows:

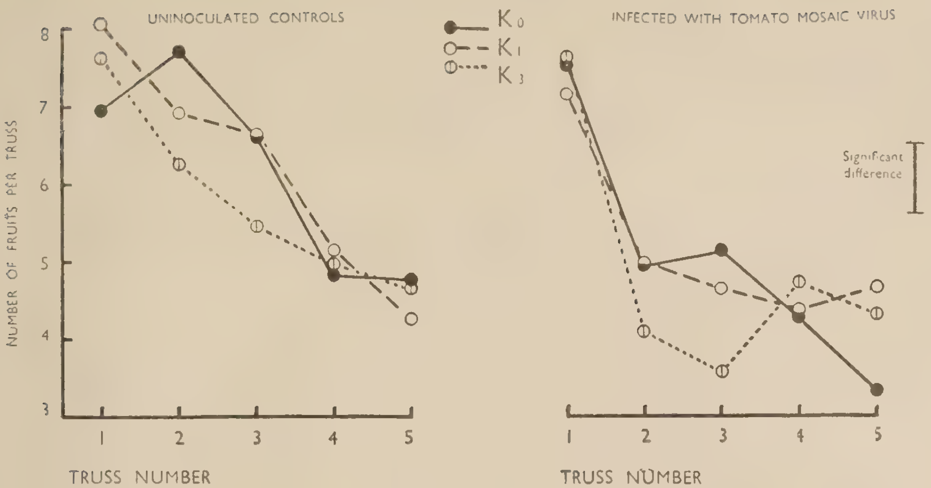
(1) *Number of flower buds.*—The "z" value for the interaction T × K × V was **0·5270** (1% point = 0·4604). The graphs in Text Fig. 3 show that significant differences between



TEXT FIG. 3.
Influence of potash manuring on number of flower buds.

control and inoculated plants occurred only at the lowest potash level (K_0), and here only in the fourth and fifth trusses. Symptoms of Mosaic infection did not begin to appear in any control plant until some days after the flower bud counts had been completed, so that it is unlikely that flower bud differentiation was affected by this factor in control plants at any manurial level. There were no leaf symptoms suggestive of potash deficiency in any of the plants of the low potash group at this stage.

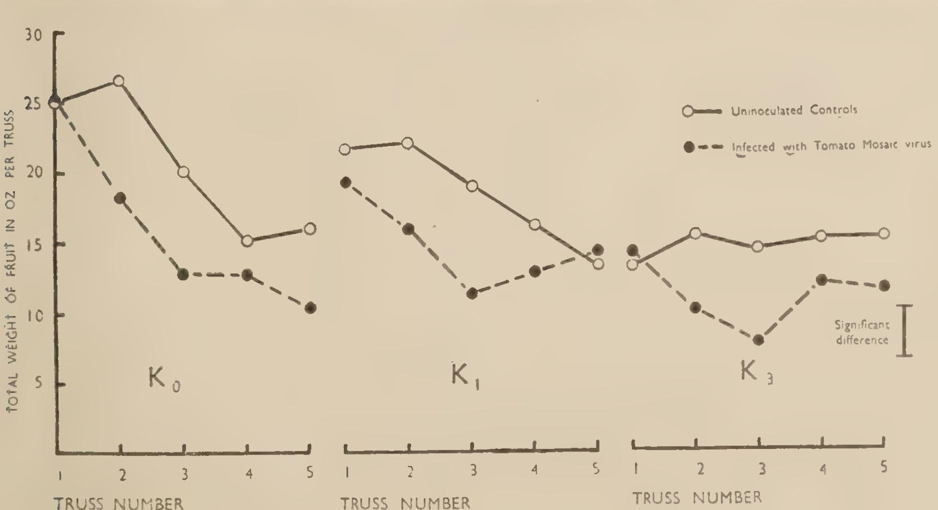
(2) *Number of fruits.*—The "z" value for the interaction T × K × V was **0·4391** (5% point = 0·3983). The following significant differences between inoculated and control plants at the three potash levels are given graphically in Text Fig. 4. A possible interpretation of these results will be discussed later.



TEXT FIG. 4.
Influence of potash manuring on number of fruits.

Truss 1.	Controls.	K_0 is lower than K_1.
	Inoculated.	No significant differences between K_0, K_1 and K_3.
Truss 2.	Controls.	No significant difference between K_3 and K_1.
	Inoculated.	K_3 is lower than K_1.
Truss 5.	Controls.	No significant difference between K_0, K_1 and K_3.
	Inoculated.	K_0 is significantly lower than K_1 or K_3.

(3) *Total fruit weight.*—In Text Fig. 5 is shown graphically the yield of fruit from the separate trusses in relation to Mosaic infection and potash manuring. Although the interaction T × K × V did not attain the conventional level of significance by the " z " test (see *Analysis of*



TEXT FIG. 5.
Influence of potash manuring on total yield of fruit.

Variance), the difference between fruit weight on the fifth truss of the control plants and on that of the inoculated plants exceeded three times the standard error of the mean only in the low potash group (K_o). This difference may presumably be related to the effects noted in the fifth truss for flower buds and fruit numbers. There is no evidence that the high proportion of control plants accidentally infected in the K_1 and K_3 series reduced the yield of fruit in the fourth and fifth trusses as compared with the K_o series, in which accidental infection was shown to have been delayed.

Significant interactions were found between potash level and truss position for various characters and these are indicated in Text Fig. 2. No significant interactions between truss position, Mosaic infection and lime level were found.

Mosaic infection and the setting of fruit.

As stated above, the effect of virus infection on the number of fruits produced was again the factor which appeared to be directly associated with the lower yield of the inoculated plants. There was no evidence to show that any of the lime, potash or standard soil treatments had caused any improvement in this respect. Counts were made of the number of chats produced. These are small, seedless, fruits not exceeding ½ oz. in weight, and are stated to be produced where pollination is faulty (Bewley, 1938). In all manurial treatments there were more chats on the inoculated plants than on the controls, and the great majority of these were found on the second and third trusses of the inoculated plants (see Fig. 5, Plate II).

An analysis of the fate of the flower buds is given in Table VI (Mean of all treatments).

TABLE VI.

Fate of the flower buds.

	No. of flower buds per plant.	No. of fruits.	No. of chats.	No. of buds or fruits abscissed (by difference).
Uninoculated controls	36·85	30·40	0·42	6·03
Inoculated with Mosaic virus	35·63	25·08	1·90	8·65

The conditions leading to the formation of chats and to flower-shedding in healthy plants may be similar, viz. low humidity, dry soil and extremes of temperature, and would appear to be related to changes in the water relations of the plants. It is probable that the formation of chats on the infected plants, particularly at the time when these plants were susceptible to wilting, is associated with the virus-induced changes in the water relations of the plants. It should be noted, however, that there was much variation in the numbers of chats produced by adjoining plants receiving similar treatments.

The effects of additional liming.

The incorporation of 2 oz. of slaked lime per pot of soil containing initially 4·39 per cent. of calcium carbonate (on dry weight basis) was found to reduce the numbers of flower buds and fruits, the average fruit weight and the total yield of fruit. The slightly alkaline reaction of the soil did not apparently affect fruit yield adversely when other factors were favourable, and the effects associated with additional liming are unlikely to be due to an unfavourable soil reaction. Only relatively small changes in pH were found to occur in the soil during the course of the experiment (see Table I).

The fact that the slaked lime was mixed in the soil immediately before planting suggested that the freshness of the lime may have adversely affected root growth. This was partly confirmed by the occurrence of wilting of the plants, thirteen days after potting. On this occasion

there was bright sunshine and all the plants were watered and damped overhead in the morning. At 3 p.m. a count was made of the numbers of plants wilting with the following results :

Treatment.	*Plants wilting.*	*Treatment.*	*Plants wilting.*
Ca_0K_0	0	Ca_2K_0	25
Ca_0K_1	0	Ca_2K_1	12
Ca_0K_3	1	Ca_2K_3	9

This effect appeared to be quite transitory, however ; for on May 5th, when plants were again wilting under similar conditions, no such association could be demonstrated, and wilting on that date was confined to plants inoculated with the Mosaic virus two weeks previously. Since no significant interaction was found between lime level and truss position for any of the characters measured, it seems reasonable to conclude that there was, in fact, a real depressing effect on plant growth due to the additional liming, which was distinct from any temporary check which may have resulted from planting in a soil to which the lime had been freshly added.

This conclusion confirms the results obtained by Bewley (1933) on the soil of this nursery, from experiments in which slaked lime was applied to soil and flooded in, not less than six weeks before planting tomatoes. The yield of tomatoes was reduced by this treatment as compared with that of plants receiving no additional lime. Lister (1916), also working with the soil of this nursery, found that liming the soil of pot plants did not greatly affect the yield of tomatoes but he obtained the highest yields from soil receiving 2·5 per cent. (of their moist weight) CaO, and it was immaterial whether chalk or caustic lime was used. In the latter work, however, there is no record of the time which elapsed between liming and planting.

It is possible that the marked reduction in total fruit yield of 18·7 per cent. (mean of all treatments) associated with additional liming, may be partly a varietal response, akin to the reaction of this variety to potash which is discussed below. Further work on this subject is desirable in view of the large applications of lime to glasshouse soils in this country.

The effect of potash manuring.

Increasing the potash level reduced the total number of flower buds produced on five trusses significantly, but had no significant effect on the total number of fruits. Omission of potash reduced the percentage of flower buds setting fruit from 77·7 per cent. in the medium potash series (K_1) to 74·1 per cent. (K_0).

The marked reduction in the total yield of fruit associated with increasing dressings of potash must be attributed primarily to the effect of potash on average fruit weight. The mean values for average fruit weight are given in Table VII together with the total yield of fruit from the potash plots.

TABLE VII.

Mean values for average and total fruit weight.

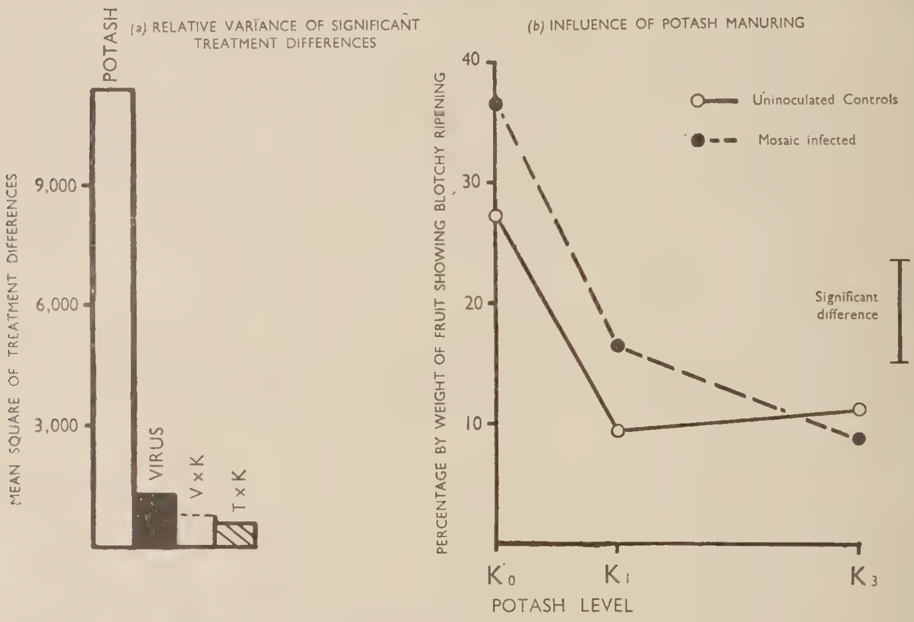
Treatment.	Average fruit weight, in oz.	Total fruit weight, lb.	oz.
K_0	3·225	5	10·75
K_1	2·884	5	2
K_3	2·448	4	0
Mean	2·852	4	15
S.E. of mean	±0·055		±2·7

In considering these results in the light of commercial practice it will first be necessary to present the data concerning the effect of potash on the incidence of blotchy ripening. It may be stated at once, however, that these results are almost certainly related to the habit of growth

of the variety Potentate, and may not apply generally to all varieties of tomato. It must also be pointed out that the initial reserves of potash in the soil were high (see Table I), and symptoms of true potash deficiency appeared only on a few plants and then only when five trusses of fruit had set. It is probable that small quantities of potash may have been obtained by roots entering the soil beneath the pots, although rooting-through did not occur in all pots and even then was very limited in extent.

Blotchy ripening.

The magnitude of the variance associated with potash level in the percentage of fruit by weight showing severe blotchy ripening is indicated graphically in Text Fig. 6 in which the mean



Text Fig. 6.

Percentage by weight of fruit showing blotchy ripening.

squares of the treatment differences are plotted in the manner described for Text Fig. 2. The variance associated with Mosaic infection was not high, relative to that associated with potash level, although it did attain the level of significance. The mean values for potash and virus treatments are given in Table VIII.

Table VIII.

Percentage of blotchy fruit (five trusses).

Treatment.		Treatment.	
K_0 K_1 K_3	31·88% 12·83% 9·85%	Uninoculated controls Inoculated	15·86% 20·51%
Mean S.E. of mean	18·19% ±1·99%		18·19% ±1·63%

The lowest mean value recorded for all treatment of the main experiment was 7·8 per cent. of blotchy fruit from inoculated plants receiving additional lime together with high potash (Ca_2K_3). This was also the treatment yielding the smallest weight of fruit (Table V) and is in agreement with the result reported previously (Selman, 1942) that the truss yielding the least weight of fruit was also the truss which produced the least percentage of blotchy fruit.

Uninoculated plants grown in the standard potting soil, which contained stable manure, produced a high total yield of fruit (Table V) but only 3·8 per cent. of blotchy fruit. It is uncertain whether this result is to be attributed to a better water supply to the fruits—the concomitant of improved soil texture leading to a more effective root system—or to be associated with an increased supply of minor elements or soluble organic nutrients. It seems possible that all these factors may have contributed to the high quality of fruit in this group of plants, and it is probable that potash level and Mosaic infection are not the only important factors controlling blotchy ripening in the tomato.

The interaction between potash level and Mosaic infection failed to reach the level of significance ("z" = 0·4354; 5% pt. = 0·5994), but is of interest in view of the interactions recorded above between potash level and Mosaic infection. The graph in Text Fig. 6 shows that increasing the potash level decreased the percentage of blotchy fruit produced by the inoculated plants, whereas a high level of potash (K_3), i.e. in excess of that normally applied in commerce, tended to increase the percentage of blotchy fruit in the uninoculated controls.

That the effect of potash on blotchy ripening is not simple, is suggested by the significant interaction found between truss position and potash level ("z" = 0·7203; 0·1% pt.=0·5917). Where potash was applied (K_1 and K_3), the percentage of blotchy fruit on the separate trusses tended to increase from the first to the fifth truss, but where potash was omitted (K_0) the proportion of blotchy fruit was found to *decrease* on ascending trusses, although the relative deficiency of potash might have been expected to increase in the higher trusses. This result may be related to the effects of potash manuring on the uptake of nitrogen by the plant and will not be further elaborated here.

Blotchy symptoms induced by shortage of potash could not usually be distinguished from those associated with Mosaic infection. Where potash shortage was a contributory factor, the symptoms were of the type shown in Fig. 6, Plate IV, in which red pigmentation always started at the stigmatic end of the fruit and spread in irregular sectors towards the calyx end. When medium potash was supplied, Mosaic infection was frequently associated with a type of blotchiness in which irregular islands of tissue failed to ripen, but in which no sectoring was apparent. Many intermediate types of fruit symptom were observed on plants from all treatments, however, and it is of interest to record that over 50 per cent. of the fruit picked from plants which had received no additional potash failed to show blotchy ripening, even when inoculated with Mosaic virus (see Table V).

DISCUSSION.

The relation between sulphate of potash and Mosaic infection under commercial conditions.

It is common nursery practice to apply about 1 ton of sulphate of potash per acre annually for the tomato crop under glass, ½ ton in the base and up to ½ ton in top dressings, irrespective of the initial potash content of the soil. In conjunction with the appropriate nitrogenous, phosphatic and organic manures this has been found to yield profitable crops of good quality fruit, when applied to the majority of commercially-grown varieties. There is, however, apparently little evidence that the weight of fruit produced by the tomato is very markedly increased by these heavy dressings of potash. Data based on ash analyses of fruit and haulm have been taken as evidence to suggest that the potash requirement of the tomato is relatively high (Lewis and Marmoy, 1939), but the mere presence of potash in the tissues is not necessarily evidence for its essentiality in these amounts.

Trials were made at this Station from 1916-21 (Bewley, 1921) comparing crop yields from plants receiving complete artificial manures with plants receiving complete artificials without potash. Two varieties of tomato were grown, Kondine Red, a variety of the Ailsa Craig type but rather less free rooting, and Comet, a variety in some respects similar to Potentate, although not so shallow-rooting as the latter. Over a five-year period with Kondine Red continuous omission of potash gave a total reduction of approximately 8·5 per cent. in total fruit weight, whereas Comet, over a *six*-year cropping period, showed a reduction in yield of only 2·3 per cent. as a result of omitting potash. In subsequent years serious losses in crop did in fact occur, as a result of a deficiency of potash in the soil, together with the typical leaf symptoms associated with deficiency of this element ; but it is clear that the level of potash necessary in the soil for satisfactory tomato production may not be the same for all varieties.

From earlier investigations at this Station and from the present experiment, it would seem that the reasons for the heavy applications of potash must be related primarily to the improved quality of fruit obtained, particularly where Mosaic infection is widespread, and secondarily to the varieties of tomato most commonly grown. For the majority of commercial varieties as Bewley (1938) has stated : " It is advantageous to apply 5 cwt. of sulphate of potash per acre 6-8 weeks after planting. . . . The dressing has the effect of steadying growth, preventing ' blotchy ripening ' and giving resistance to ' Stripe disease.' " The variety Potentate would appear to respond better to rather different treatment, cropping well in the warm, humid conditions of the cucumber house and making unnecessary the potash-induced check to vegetative growth known to be so valuable in facilitating fruit set in the more vigorous-rooting varieties.

Where Mosaic infection is widespread, the results of the present work would indicate that free application of sulphate of potash improves the quality, but not the quantity of the fruit. In recent years, however, determined efforts have been made, under commercial conditions, to grow tomato crops free from virus disease, by the use of virus-free seed and the application of other control measures. When this is done it may be suggested that the health of the plants can best be maintained by restricting the potash supply in the soil to a relatively low level, applying enough to supply basic growth requirements, but not quantities sufficient to check growth and thereby increasing the susceptibility of the plants to accidental infection with Mosaic. If at the same time adequate supplies of suitable organic materials be maintained in the soil, blotchy ripening should be reduced to negligible proportions, and high total yields be maintained.

The relation of potash to number and quality of fruits produced by Mosaic infected plants.

There is no evidence of a significant interaction between potash level and Mosaic infection for the total crop produced by five trusses ; the effect of such infection on the flowering and fruiting of the separate trusses may, however, be determined by the level of potash manuring.

(*a*) *Flower buds.*—There was a reduction in the numbers of flower buds produced by infected plants, but only where the potash was tending to a low level in the soil (i.e. on trusses 4 and 5 of the K_0 series).

(*b*) *Number of fruits.*—In the *first* truss of the controls, omission of potash was associated with significantly fewer fruits than medium potash manuring (K_1). This result is paralleled by the early dressings of potash, recommended by Bewley, in order to steady growth and facilitate fruit set in the first truss. The control plants receiving no potash were in fact soft and leafy at this stage (Fig. 1, Plate I) in contrast to plants receiving high potash (Fig. 2, Plate I). On the other hand, with Mosaic infection, there was no reduction in the numbers of fruits produced on the plants without potash. Now Mosaic infection before symptom appearance, tends to reduce the percentage water content of the plant ; and this reduction reaches a maximum a few days after the appearance of symptoms (Ainsworth and Selman, 1936), after which time the water content tends to rise above that of healthy controls. Thus, it might appear that

during the setting of the first truss on the inoculated plants without potash, the hardening action of the virus disease induced an effect analogous to that which potash application induced on healthy plants.

When the flowers of the *second* truss were setting on the inoculated plants, the plants were at the point of maximum reduction of water content and also one at which susceptibility to wilting was most marked. Hence excessive potash manuring might be expected to harden the infected plants to an even greater degree, with a resultant reduction in the number of fruits setting on plants receiving K_3 relative to those receiving K_0 or K_1, a difference which was smaller and non-significant in the control plants, which had suffered only the potash-induced hardening effect.

In the *third* and *fourth* trusses no significant interactions were found, and this may be associated with the secondary increment in percentage water content or softening of the infected plants, so that potash manuring was not detrimental to fruit set in the infected plants at this stage.

With the flowering of the *fifth* truss in the K_0 series, the level of potash was tending to a low value in the soil, and the effect of this factor on flower bud production in the infected plants was presumably reflected in a reduction in the numbers of fruits produced on this truss, an effect not noted with the controls.

(*c*) *Percentage of blotchy fruit.*—As has been stated, the incidence of blotchy ripening was not affected in identical manner on control and infected plants when the heaviest dressing of potash was applied. This result may be related in part to the large amount of accidental infection which occurred in this group of control plants. It may also be related to the effect of high potash manuring on the water relations of the plant and of the fruit.

There is thus evidence to suggest that the potash requirement of the tomato crop may to some extent be determined by the health or Mosaic infection of the plants, both in relation to the flower and fruit production of certain trusses and also to the production of high quality fruit. The application of potash for the control of Stripe or Streak symptoms in Mosaic infected tomato plants, has been successful under commercial conditions (see Bewley, 1938). The response of potato plants affected with Leaf Roll to potassium nitrate at two different temperatures was found to differ from that of healthy controls (Butler and Murray, 1932). It is evident, therefore, that under certain conditions, potash manuring may be a factor of considerable value in the successful culture of virus infected plants.

Susceptibility to accidental infection.

Of considerably more practical importance than the influence of potash manuring on Mosaic infected plants is the relation of potash manuring to the incidence of Mosaic infection in the uninoculated control plants. Further work is necessary to determine the nature of this effect. It was, however, noted that plants grown without additional sulphate of potash made freer and more leafy growth than did plants receiving potash, particularly in the earlier phases of growth. Other workers have recorded a similar relationship between the incidence of virus disease and the growth of the plant. Thus, Curtis (1940) noted that where bed-sown tobacco plants were used for transplanting (i.e. presumably where the check to growth of the transplant was least) the amount of initial Mosaic infection was negligible. This relative freedom from Mosaic was associated with soil and moisture conditions favourable for the tobacco plants.

Crowther (1941), studying seasonal growth and yield of cotton in the Sudan, was led to conclude that although the Leaf-Curl disease (and also Blackarm) was severe in years of low yield, the growth data collected early in the season proved that growth was inferior *before* the disease had become widespread. The factor believed to be responsible for seasonal fluctuation in growth and yield was a soil factor with which the amount of nitrogen available for the crop was closely associated. Soil conditions conducive to vigorous growth were thus associated with a reduced incidence of virus disease.

The important findings of Merkenschlager and Wartenberg (1931) on degeneration in the potato provide further evidence of the rôle of potash manuring in determining the health of the plant. Normal plants grown in sandy soils heavily manured with potash, produced tubers with the typical composition of diseased specimens, and the resulting plants displayed the usual characteristics of Leaf Roll. It was concluded that the limit of total ash and K_2O in seed tubers which permits healthy growth of the succeeding crop appears to be rather sharply defined. Wartenberg *et al.* (1935) were led to conclude that two factors contributed to the running out of potatoes. The primary one was considered to be a physiological disorder induced by environnental conditions to which the variety is not well adapted, and this creates a predisposition to infection by the various virus complexes which constitute the second factor. It may be suggested from the present work that these two factors are similarly operative in reducing the yield of the tomato crop, although the perpetuation of the disorders is effected differently. Vegetative propagation is rarely practised with the tomato, and seed transmission of the tomato Mosaic virus may occur only in a relatively small proportion of the seedling population. Nevertheless, the ease with which the virus may be spread from plant to plant by man's activities may rapidly lead to a high incidence of infection, particularly where certain environmental conditions tend to inhibit the free growth of the plants.

It should be noted that not all environmental factors tending to check plant growth cause an increase in the susceptibility of plants to accidental infection. Thus, additional liming was not found to produce this effect, although vegetative growth and yield were reduced thereby. With the Spotted Wilt virus there is some evidence that additional liming may decrease the susceptibility of seedling tomato plants to artificial infection (Selman, 1941*b*).

Two effects have been noted in the present work which should be clearly distinguished. Firstly, in plants *heavily inoculated* with tomato Mosaic virus there was a negative correlation between growth rate and the rate of appearance of symptoms, a fact established for tomato seedlings by Ainsworth and Selman (1936). On the other hand, uninoculated plants making the most rapid and vigorous growth tended to remain relatively immune from accidental infection with this virus. It might appear therefore, that in plants of a certain type, or at a certain stage of development, the introduction of small numbers of virus particles may result in either a failure of the virus to multiply or to be released from the site of entry. Samuel (1934) found that in pot-grown, fruiting, tomato plants, complete invasion with the tobacco Mosaic virus may require two months when a single leaflet is inoculated. He also demonstrated that inoculation of large field-grown tomato and tobacco plants did not, in two months, result in complete invasion of mature leaves with the virus.

It is possible, therefore, that where growth was unrestricted by excessive potash manuring, the leaves attained maturity (although not necessarily senility) more rapidly than those of plants receiving large quantities of potash, and that this led to some localization of virus particles introduced accidentally during cultural operations.

SUMMARY.

1. Flower and fruit production in tomato plants, var. Potentate, inoculated with tomato Mosaic virus (A.17) when the first truss was in bloom, has been compared with that of uninoculated controls, the differences being studied at two levels of lime and three levels of potash manuring, selected in close relation to those employed in commercial practice. The steam sterilized soil was an old clay-loam containing reserves of both lime and potash, and the manures used were dried blood, superphosphate, slaked lime and sulphate of potash. The plants were grown in 10-inch pots under glass, and five fruit trusses were allowed to develop.

2. The appearance of disease symptoms in the inoculated plants was delayed where growth was retarded by lime and potash applications. Severe yellow-green foliar mottle was confined to the most vigorous plants grown without additional lime.

PLATE I.

Fig. 1.

Uninoculated control plant of Ca_0K_0 series.

Fig. 2.

Uninoculated control plant of Ca_2K_3 series.

Photo: April 30th. Illustrating different types of growth three weeks after planting in ten-inch pots.
(*Reproduced on the same scale.*)

Fig. 3.

Uninoculated controls with five trusses set.

Photo: July 23rd.

[*face p.* 104

FIG. 4.
Plants inoculated with tomato Mosaic virus when the first truss was blooming, April 20th.
Photo: July 23rd.

FIG. 5.
Inoculated plants of the Ca_2K_1 series showing chats on the second truss. Trusses 1 and 3 produced good fruit.
Photo: August 24th.

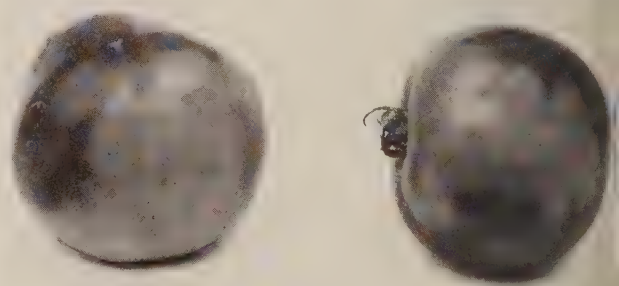


FIG. 6.
Fruits showing blotchy ripening; taken from plants grown withou[t] additional potash.
Photo: August 8th, with Trigreen filter.

3. Despite precautions, Mosaic symptoms began to appear on the control plants seven weeks after introducing virus into the glass house. Four weeks later, all the uninoculated plants were tested for the presence of the virus. Where sulphate of potash had not been applied, the number of virus-free plants was significantly greater than where it had been applied. It is suggested that the hardening or growth check, commonly associated with potash applications in commercial tomato culture, tended to increase the susceptibility of the plants to accidental infection. Additional liming did not induce a similar effect, although it tended to reduce vegetative growth and fruit yield.

4. Data were recorded for each of the five trusses of the plants separately, and included numbers of flower buds, fruits and chats together with the weight and quality of the fruit. A statistical examination of the results was made.

5. The total number of flower buds per plant was not affected by Mosaic infection, but it was reduced by liming and by the addition of potash. The number of flower buds on individual trusses was related to the position of the truss.

6. The total number of fruits was reduced by Mosaic infection and by liming, but was unaffected by the level of potash. On single trusses the number of fruits was closely related to the position of the truss, the maximum being produced on the first truss and the least on the fifth.

7. The average fruit weight was reduced by increasing the potash level, and there was a slight reduction associated with Mosaic infection and with the addition of lime. The average fruit weight of individual trusses was related to the position of the truss.

8. The total weight of fruit was reduced by Mosaic infection, by additional liming and by sulphate of potash. The total weight of fruit from separate trusses was related to the position of the truss.

9. The highest total yield of fruit was 7 lb. 0 oz. per plant, from uninoculated plants receiving neither additional lime nor potash. Of this total, 4 lb. $8\frac{1}{2}$ oz. consisted of ripe, unblemished fruit. Mosaic infected plants receiving similar manurial treatment yielded 5 lb. 6 oz. per plant, of which 2 lb. $10\frac{1}{2}$ oz. was unblemished ripe fruit.

10. Inoculated plants produced a greater number of chats than did the control plants, a result considered to be related to virus-induced changes in the water relations of the plants.

11. Mosaic infection increased the percentage of fruit showing severe blotchy ripening. Omission of potash induced a much greater increase. A subsidiary experiment indicated that the addition of stable manure and potash to soil reduced the proportion of blotchy fruit to a value below that to which it could be reduced by potash alone.

12. Total flower and fruit production was found to be affected similarly by Mosaic infection at all levels of lime and potash. Significant interactions were, however, found between truss position, potash level and Mosaic infection for flower bud and fruit numbers. It is concluded that at certain stages of development there are differences in the potash requirement of healthy and Mosaic infected plants.

13. There was evidence of differing responses to potash manuring in the amounts of blotchy fruit produced by infected and by control plants. High potash manuring tended to reduce the incidence of blotchy ripening in infected plants, whereas similar quantities of potash applied to the controls tended to increase the percentage of blotchy fruit relative to that produced by medium potash manuring of similar plants.

14. The practical aspects of potash manuring in relation to Mosaic infection are discussed. It is concluded that with the variety Potentate, potash manuring should be carefully controlled if immunity from accidental infection is to be maintained. Where bulky organic manures are not used, lighter potash dressings are likely to increase the total yield of fruit of both healthy and Mosaic infected plants, but may tend to produce fruit of inferior quality.

15. It is emphasized that the results have been obtained with the variety Potentate only. Other work at this Station has suggested that other varieties may respond differently, particularly in relation to potash manuring.

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THE ORDER AND PERIOD OF BLOSSOMING IN PEAR VARIETIES

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DURING the past four years, 1939-42, I have recorded the flowering times of varieties of pears grown at the John Innes Horticultural Institution. The trees from which the records were taken are now twelve years old and trained as cordons. They are all on Malling Quince A (Angers Quince) rootstock, and have received the same cultural and manurial treatment.

The results are given in Table I, in the first two columns of which the varieties are grouped according to the times of full bloom. These times were taken when 50 per cent. of the flowers were fully open. In the last column the duration of the flowering period is given for each variety. It was found that although the date of flowering and the length of time a variety remained in flower varied from year to year, the relative flowering times of the varieties were very consistent.

The variety Brockworth Park flowers exceptionally early and is very variable. In 1939 it began to flower on March 1st, reached full bloom on March 15th and continued to flower until April 12th, a total of 42 days. In 1940 it started to flower on April 7th and continued until April 28th. In 1941 the flowering period was from April 17th to 30th, and in 1942 from April 11th to 26th. Owing to the variability of this variety, which began to flower 26, 8, 5 and 3 days respectively, before any other variety in the four years during which records were taken, the more constant variety Zoé was taken as the standard of earliness.

The length of time during which the trees remained in flower was taken from the date of the opening of the first flower until petal fall, i.e. when 90 per cent. of the petals had fallen. The average time for all varieties was 12 days, and variation between varieties was not very great. With the exception of Brockworth Park, which flowered for an average of 21 days, the longest flowering varieties were Van Mons, Léon le Clerc, Beurré Brown and Beurré Fouqueray which flowered for an average of 16 days each. The shortest flowering period was that of four varieties which remained in flower for only ten days. The seasonal variation, however, was more pronounced; in 1939 the average flowering period was 12·1 days, in 1940 9·2 days, in 1941 15 days, and in 1942 10·7 days. There were, however, neither such wide differences nor any change in the order of flowering as occur in apples (Brown, 1940).

Biennial flowering was very marked in some varieties, particularly in Fondante de Thirriott, Joséphine de Malines, Souvenir de Congress, Grégoire Bourdillon and Michaelmas Nelis. The behaviour of these varieties should be borne in mind when interplanting, as biennial flowerers cannot be relied upon as effective pollinators.

The season of flowering is not so prolonged in pears as it is in apples, and in years when varieties flower in quick succession the flowering times of most varieties of pears overlap sufficiently for cross pollination to take place. In other years, however, when the flowering period is spread over a longer period, early and late varieties do not overlap sufficiently to provide adequately for cross-pollination. Long flowering period is correlated with low temperatures, and since pollinating insects are less active in cold than in warm weather, it is important that varieties whose flowering times closely correspond should be planted together.

In Table I the chromosome number, where known, has been put in brackets after the name of the variety. The triploid varieties (51 chromosomes) produce very little good pollen, and this should be considered when arranging the layout of plantations and gardens. The variety Marguerite Marrilat, although a diploid, is male sterile and is therefore entirely useless as a pollinator.

TABLE I.

Order of flowering based on the average of four years, 1939-42.

Order of full bloom in days.	Variety.	No. of days between 1st flower and petal fall.
−10	Brockworth Park	21
1	Zoé	13
2	Bellisime d'Hiver	14
	Beurré Giffard (34)	13
	Marguerite Marrilat (34)	15
	St. Luke (34)	11
	Van Mons Léon le Clerc (34)	16
	Vicar of Winkfield (51)	13
3	Beurré d'Amanlis (51)	14
	Doyenné d'Été	12
	Louise Bonne of Jersey (34)	11
	Passe Crassane (34)	12
	Princess	15
	Winter Orange	13
4	Baronne de Mello (34)	12
	Beurré Brown (34)	16
	Beurré Diel (51)	11
	Doyenné Boussoch (51)	11
	Durondeau (34)	12
	Fondante de Thirriott (34)	11
	Williams' Bon Chrétien (34)	11
5	Beurré de Jonghe	14
	Duchesse de Bordeaux	13
	Gansell's Bergamotte (34)	11
	Hacon's Incomparable	11
	Josephine de Malines	11
	Le Lectier (34)	11
	Petite Marguerite	11
	Thompson's	12
	Verulam	12
6	Beurré Fouqueray (34)	16
	Beurré Six (34)	12
	Chalk (34)	12
	Conference (34)	13
	Emile d'Heyst (34)	10
	Gilogil	10
	Huyshe's Prince Consort	13
	Pitmaston Duchess (51)	12
	Souvenir de Congress (34)	10
	Summer Beurré d'Arenberg	10
	Windsor	13
7	Beurré Hardy (34)	14
	Dr. Jules Guyot (34)	11
	Fertility (34)	12
	Marie Louise d'Uccle	11
	Napoleon (34)	11
8	Belle Julie	13
	Beurré Alexandre Lucas (51)	13
	Clapp's Favourite (34)	11
	Doyenné du Comice (34)	12
	Glou Morceau (34)	13
	Grégoire Bourdillon	12
	Nouveau Poiteau (34)	12
9	Beurré Mortillet (34)	10
	Catillac (51)	11
11	Michaelmas Nelis	11

TEMPERATURE AND DATE OF FLOWERING.

There is considerable variation in the dates of flowering of pears from year to year as shown in Fig. 1. This shows the number of varieties in full flower on each day during the flowering period. The variety Brockworth Park is omitted. In 1939 the first variety came into full bloom on April 10th, and all had reached full bloom by April 21st, whereas in the years 1940 and 1942 no variety reached full bloom until April 21st ; in 1941 not until April 26th.

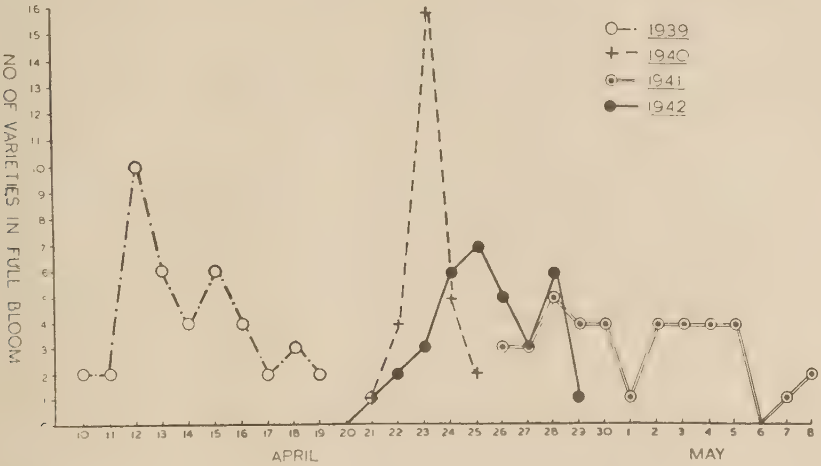

FIG. 1.

Variation in dates of flowering of pears from year to year.

Table II gives the mean temperatures (°F.) for the months before flowering, and it will be seen that weather conditions affect the time of flowering. Herbst and Meyer (1940) have studied this problem and conclude that in pears there is a threshold temperature below which flower development does not proceed, and that the effect of temperature above this threshold value, between mid-winter and flowering time, is cumulative. This threshold temperature was calculated and given as 6° C. (43° F.).

The temperature records at Merton did not include the details necessary to calculate the time in degree-hours, but from Table II it will be seen that in 1939, when the trees flowered very early, the mean temperature exceeded 43° F. in five half-monthly periods before flowering

TABLE II.

Mean Temperatures (°F.).

Time.		1938–39.	1939–40.	1940–41.	1941–42.
December	1–15	47·3	43·0	40·2	45·2
	16–31	34·5	31·8	38·1	37·9
January	1–15	42·0	31·5	32·9	34·9
	16-31	44·3	28·6	37·5	32·3
February	1–15	43·7	34·3	40·5	33·3
	16–28	42·5	43·4*	38·4	34·1
March	1–15	45·4	41·7	42·6	38·8
	16–31	41·4	46·6	42·1	45·7
April	1–15	50·2	46·2	44·0	50·7
	16-30	47·2	53·3	46·1	50·8
May	1–15	—	—	41·9	—

* February 16–29.

began. On the other hand, in the year of latest flowering (1941), the mean temperature did not exceed 43° F. until the first half of April. It therefore seems probable that here also the development of flowers proceeds only at temperatures above 43° F.

SUMMARY.

1. The order and period of blossoming of 56 varieties of pears on Malling Quince A root-stock is given from records taken in the years 1939-1942.

2. It was found that varieties came into flower in approximately the same order each year, but the date when they began to flower varied from year to year. The average number of days between flower opening and petal fall also varied, being 12·1, 9·2, 15·0 and 10·7 respectively in the four years mentioned.

3. In the short-flowering years, 1940 and 1942, some varieties overlapped enough for effective cross-pollination to occur, but they did not do so in the long-flowering years 1939 and 1941. To allow for these year-to-year differences it is important therefore to interplant those varieties whose flowering times most closely correspond.

4. Differences in temperature affect the time of flowering, and it appears that flower development proceeds only at temperatures above 43° F. (6° C.).

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THE STATISTICAL INTERPRETATION OF VIGOUR MEASUREMENTS OF FRUIT TREES

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Introduction.

In subjecting the results of biological experiments to statistical analysis research workers are sometimes faced with difficulties, for when statisticians are devising methods they are apt to make assumptions for the sake of simplicity that lead to their work being inapplicable in certain fields of research. Of late years, at East Malling, the suspicion has been growing that the usual methods of analysis are not always suitable for dealing with vigour records of fruit trees, and an example can easily be suggested. Consider an experiment designed to compare the vigour of apple trees on a series of rootstocks ranging from the dwarfing Malling IX to the very vigorous Malling XII, the trees on them being arranged in randomized blocks. The results would ordinarily be examined by the method of the analysis of variance. As a result a single standard error, applicable to the whole experiment, would be calculated ; and from this the significance of the difference between any two rootstocks would be judged. The validity of such a procedure plainly depends on the accuracy of the assumption that there is, in fact, a single standard error applicable to the whole experiment, i.e. that trees are equally variable on all rootstocks. Anyone who has dealt with sets of trees knows that, in absolute measurements, trees on a vigorous rootstock show a wider range of variation than those on a dwarfing one, though proportionately the variabilities might be about equal. Thus it appears likely that a single estimate of the standard error will give undue significance to differences at the top of the scale and belittle real differences at the bottom.

If the variation, as measured by the variance, is, in fact, dependent on the mean value of the observations being dealt with, the difficulty is one that has been met with in other fields and has been successfully overcome. The usual procedure is to " transform " the data, that is to say, to analyse some function of them that is statistically unobjectionable, while using the untransformed data for the statement of results. Thus, if the variances are proportional to the mean it is best to carry out the analysis on the square roots of the original observations. Bartlett (1936) adopted this expedient in analysing the numbers of cockchafer larvae in a standard area, while, in dealing with counts of the aphis *Capitophorus fragariae* on strawberry plants, Greenslade and Pearce (1940) found it necessary to use the transformation $\sqrt{n+\frac{1}{2}}$, which also had been suggested by Bartlett. If the variance is proportional to the square of the mean, Cochran (1938) advised the use of logarithms. Bliss (1937, 1938) recommended the use of an angular transformation for analysing data in the form of percentages and Beall (1941-42) developed another transformation for use with certain classes of insect counts. Thus, this is no new problem, and many kinds of data have been found with this abnormality of being unequally variable, thus calling for a modification of the original analysis of variance.

It might be objected that in the present state of horticultural knowledge the differences between treatments under investigation are not large enough for this abnormality to matter, even if it exists. Suppose, to take an extreme example, that the variance is proportional to the square of the mean ; then, if a proposed manurial treatment will increase vigour by 10 per cent., the variance for this treatment will be only 21 per cent. greater than that for the control ; and such a discrepancy is of little importance. It is true that most of the large effects were investigated in the days when it was usual to calculate a separate error for each comparison and when, therefore, the difficulty had not arisen ; but even now it is not unusual for a new treatment to be unexpectedly efficacious, and sometimes large differences are introduced deliberately. Thus, widely different varieties on a range of rootstocks might well be included in a pruning experiment,

despite the likelihood of obtaining extremes of vigour. It follows, therefore, that if the variability of vigour measurements is not constant over the range of possible values, it is necessary to know which is the best transformation.

It is the purpose of the present paper to examine some of the many vigour measurements that have been employed to determine the sizes of trees. Probably the best measure, though obviously only a final one, is the total weight. During the life of the tree other characteristics must be measured, such as annual extension growth, weight of wood removed in pruning, girth of stem and height and spread of head ; and of these the most valuable are the total extension growth and area of cross-section of stem as calculated from the girth ; for it is the last two that most nearly reflect differences in tree weight. The work presented below examines these three measurements in order to judge whether their variances are independent of tree size, and a note is added on the form of their distribution. Where the original measurement is shown to be unsatisfactory, recommendations are made as to the better transformation to use, namely the square root or the logarithmic one.

Statistical method.

The procedure throughout will be to use several series of trees comparable in all respects but for their vigour, and to work out the variance for each series. The set of variances thus obtained will be tested for homogeniety by the quantity, L_1, which has been tabulated by Nayer (1936). This has a maximum value of unity when all the variances are equal, and becomes smaller as heterogeniety increases.

The word " significant " is to be taken to imply the significance level, $P = 0{\cdot}05$, unless otherwise stated.

Experimental material.

Data were examined from four trials.

I. This trial involved 32 pairs of trees, 16 pairs having the apple rootstock Malling IX and 16 Malling XII, both as rootstock and scion. For each variety half the pairs were pruned and half unpruned. All the variances given in Table I refer to variation between trees of a pair.

Table I.

Means and variances of tree weight and extension growth and their transformed values for pruned and unpruned trees of two apple varieties.

Variate.			Unpruned.			Pruned.		
			Malling IX.	Malling XII.	L_1.	Malling IX.	Malling XII.	L_1.
Tree weight after 7 years. (Pounds)	*Actual.*	Mean	9·50	76·06		4·12	42·69	
		Var.	4·50	314·94	0·236†	0·12‡	31·44	
	Sq. root.	Mean	3·02	8·66		1·97	6·51	
		Var.	0·121	1·087	0·600†	0·005‡	0·211	
	log.	Mean	0·95	1·87		0·59	1·62	
		Var.	0·0111	0·0118	0·9996	0·0012‡	0·0035	
Extension growth after 6 years. (Metres.)	*Actual.*	Mean	32·4	159·4		19·5	143·6	
		Var.	109	671	0·692*	14	632	0·289*
	Sq. root.	Mean	5·60	12·51		4·38	11·94	
		Var.	0·683	1·194	0·963	0·198	0·992	0·745*
	log.	Mean	1·49	2·19		1·28	2·15	
		Var.	0·0145	0·0065	0·925	0·0113	0·0069	0·970

* Significant, P <0·05. † Significant, P <0·01.

‡ Result vitiated because the unit of measurement was too large.

II. Use was made of records from sixteen trees of Lane's Prince Albert apple on each of the rootstocks Malling IX, II, I and XII, which show a wide range of vigour. Each group of sixteen trees was divided into two blocks of eight, and in each block the fruit of half the trees had been thinned and half not thinned. The variances given in Table II are those due to error after the elimination of block and thinning effects.

TABLE II.

Means and variances of extension growth and area of cross-section of stem together with their transformed values for Lane's Prince Albert on four rootstocks.

Variate.			Rootstocks.				L_1.
			Malling IX.	Malling II.	Malling I.	Malling XII.	
Extension growth in 7 years. (*Metres.*)	*Actual.*	Mean	117	288	391	623	
		Var.	943	8,992	6,275	8,433	0·759†
	Sq. root.	Mean	10·7	16·7	19·7	24·9	
		Var.	2·04	8·71	4·16	3·44	0·870
	log.	Mean	2·05	2·43	2·58	2·79	
		Var.	0·0143	0·0269	0·0083	0·0043	0·805*
Area after 19 years. (*Sq. cm.*)		Mean	50	239	277	370	
		Var.	92	3,123	2,100	1,508	0·573†
Girth after 19 years. (*Millimetres.*)		Mean	250	545	588	681	
		Var.	600	4,438	2,508	1,285	0·775†
log. (Area).		Mean	1·69	2·37	2·44	2·57	
		Var.	0·0075	0·0128	0·0059	0·0020	0·824*

* Significant, P <0·05. † Significant, P <0·01.

III. Thirty-one pairs of Cox's Orange Pippin on Malling IX, which had received adequate potash since planting, were compared with thirty pairs which had been potash starved and were consequently stunted. Extension growth was recorded on only sixteen pairs of each kind. All variances given in Table III are between trees of a pair.

TABLE III.

Means and variances of extension growth and area of cross-section of stem together with their transformed values for Cox's Orange Pippin on two manurial treatments.

Variate.		Low Potash.		High Potash.		L_1.
		Mean.	Var.	Mean.	Var.	
Extension growth in 8 years (*Metres.*)	*Actual*	56·6	326	133·0	2,176	0·673*
	Sq. root	7·40	1·62	11·34	4·60	0·878
	log.	1·83	0·0311	2·09	0·0334	0·999
Area after 14 years. (*Sq. cm.*)		16·3	22·0	31·0	77·0	0·831*
Girth after 14 years. (*Millimetres.*)		141	401	194	764	0·950
log. (Area).		1·20	0·0192	1·47	0·0162	0·996

* Significant, P <0·01.

IV. Records from twenty-four plots of Cox's Orange Pippin were used, each containing four trees. Four plots were included on each of six rootstocks giving widely different performances. Variances were worked out between similar trees within a plot.

EXAMINATION OF RESULTS

Weight of tree.—It will be seen from Table I that the unpruned trees of Malling IX and XII, which differed widely in their mean weight, had very unequal variances also. Transforming

to square roots caused an improvement, but when the logarithmic transformation was used the variances were nearly equal. Unfortunately the results from the pruned trees were of little use, because weighing to the nearest pound did not show any differences at all between most of the trees on Malling IX ; but the logarithms would again seem to have been best. Variances were worked out also between trees within plots on Trial IV, and when grouped into sets of six, one variance to each stock, gave L_1 values of 0·872, 0·967 and 0·949 for the tree weight, its square root and its logarithm respectively. Although none of these values differs significantly from unity, it would seem that both transformations gave some improvement, and, with the results of Trial I, indicate that the logarithmic one is probably to be preferred.

Extension Growth.—Table I shows that the untransformed variate gave widely different variances with the two varieties. Both the square root and logarithmic transformations gave marked improvement for the unpruned trees, while for the pruned trees the logarithms were much the better. Table II would suggest a preference for the square root transformation, but in Trial III, as shown in Table III, the logarithmic one proved highly satisfactory. From these results the logarithmic transformation is recommended.

Area of cross-section of stem.—Table II shows the logarithmic transformation to have been the best of the three variables; for, though it gave a significantly heterogeneous set of variances, they do not appear to be correlated with the means. This result is confirmed by Table III. Consequently it is recommended that cross-section be analysed in logarithmic transformation when the differences in treatment means warrant the extra labour. It may be noted that log (area) $= 2 \, log$ (girth) $- \, log \, 4\pi$, and that consequently the logarithm of girth would be equally suitable.

Normality of distribution of the measurements considered.—The distribution of a variable quantity can take on many forms. Thus, there may be a tendency for values to cluster below the mean and for this to be balanced by a few very high values. Sometimes, too, the values occur fairly thickly inside a certain sub-range of the whole but are sparse outside, while at other times the main body of values round the mean thins out gradually. In the analysis of variance it is assumed that the distribution is of the so-called Normal or Gaussian form, and this assumption, too, requires checking for the measurements here considered. It has, indeed, been shown by Pearson (1931) and by Eden and Yates (1933) that non-normality of distribution does not lead to an unusual number of differences being judged significant when they are really due to chance, but it is still not clear whether it may not lead to differences being missed that are in fact genuine. Also non-normality is often associated with dependence of the variance on the mean, the first property examined.

The data used for this subsidiary investigation were (*a*) those published by Knight and Hoblyn (1934) for two-year-old Grenadier apple trees on Malling VI, (*b*) those concerning fifty-four two-year-old trees of Williams' Bon Chrétien pear on Quince A, an incompatible combination. The results may briefly be summarized thus : the weight of a tree, whether of the top only, or of the root system only, or of the whole, appeared to be normally distributed. For the pears the non-normality of the logarithms was not serious but for the apples this point could not be investigated. No data were available for extension growth. For area of cross-section of stem the distribution was significantly asymmetrical, but the logarithms were nearly normally distributed, only the pear data being available. These investigations were carried out using the method of Fisher (1938). It seems unlikely, then, that the form of distribution of the logarithms of these measurements would be a source of unreliability.

DISCUSSION.

It is clear from the tables here presented that where varieties or treatments in an experiment give trees widely different in vigour, large differences in the absolute variation of those trees must also be expected.

To analyse the original measurements by any method that gives a single estimate of error would therefore be wrong. The somewhat laborious procedure of examining the results in two or even more parts might be attempted ; but, apart from the difficulties, some information would inevitably be lost. To analyse the results of such a trial as a whole it is necessary to be assured that the variate is suitable, and the work presented above affords grounds for believing that the logarithms of tree weight, extension growth and either girth or area of cross-section of stem are available for use in this way. Of course, a significant interaction obtained with the logarithms of a variate does not mean the same as if it had been obtained with the original data. It means, not that it is necessary to reject the hypothesis that the treatments have in all circumstances had the same *absolute* effect, but that they cannot have had a constant *proportional* effect ; however, with large differences this is often the more useful concept. For no measurement was the logarithmic transformation worse than the untransformed variate, and usually it was considerably better.

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SUMMARY.

Evidence is given to show that in applying the analysis of variance to measurements of tree weight, extension growth, and either girth or area of cross-section of stem of fruit trees, it is better to use the logarithms of the data. This result was arrived at by consideration of the stability of the variances with different means, and the normality of the distribution of these measurements.

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FURTHER STUDIES ON NEW VARIETIES OF APPLE ROOTSTOCKS

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In 1933 full descriptions were published of a series of families of apple rootstocks raised from crosses between the Malling selections of so-called Paradise apples (Tydeman, 1933). All the nineteen families involved in this study had one common parent, Jaune de Metz (Malling No. IX), while, included among the other parents were practically all the remaining Malling varieties of Paradise. The families were bred by the writer at East Malling in 1929.

Studies on a family of seedlings from a cross made by Crane at Merton between Malling Nos. VIII and IX were already revealing that these two rootstocks, the most dwarfing of the whole Malling series in their influence on the scion, had given rise to seedlings whose influence on the scion ranged from extremely dwarfing to extremely vigorous (Tydeman, 1937). There seemed some evidence, too, that the union of these two rootstocks had transmitted the character of early blossom- and fruit-production, not only to the more dwarfing of these seedling rootstocks but to the more vigorous ones as well. These experiments were not continued beyond the sixth year, and it is not known how far the early heavy blossom-production of the trees on the more vigorous rootstocks would have modified their growth in later years. Genetically, these results were thought to be due to the existence of complementary factors for vigour, borne separately by the dwarfing rootstocks, and it became of interest to follow this initial crossing between the more dwarfing varieties with others between varieties showing a different scion influence.

Since the more immediate practical aim in these experiments was the production of apple rootstocks possessing new combinations of economic value, in describing and classifying these seedlings special emphasis was laid from the first on such characters as ease of vegetative propagation, freedom from disease and absence of lateral spines, although in one season a fairly elaborate biometrical study of the inheritance of leaf shape and petiole length was undertaken (Tydeman, 1935).

From 1930 until 1934 approximately one thousand seedlings were grown as stools. They showed wide variation in the ease with which they produced adventitious roots and in other rootstock characteristics. Considerations of available space made it necessary greatly to reduce the numbers before proceeding further and, on the basis of their early nursery behaviour, it was possible to select thirty-eight individuals which had shown outstanding promise. The parentage of these selected stocks may be seen in Table I.

During 1935 all the suitable stocks of the thirty-eight selected seedlings were budded with Lane's Prince Albert for a preliminary trial of their influence on the scion. The unit of trees ultimately available varied considerably, and trees on Malling Nos. IX and XII were included as controls. Since only an indication of the effect of these stocks could be expected from the numbers of trees included, and as it was planned to follow this trial with a more elaborate one when sufficient material had become available, only single blocks of variable size of trees on each rootstock could be compared. The stocks were planted at about 3 feet apart in two rows 7 feet apart and remained growing where they were budded. The maidens were left to grow on naturally as feathered trees and, apart from the removal of very low branches which interfered with cultivation, the trees were not pruned in any way. Records of wood growth, blossom formation and fruit production were made until the summer of 1939 when the trial was discontinued.

A further set of trees on these rootstocks was budded during the summer of 1939. The scion variety then was Cox's Orange Pippin, because this variety with its more erect and compact

Table I.

Details of wood growth, blossom formation and fruit production. Cox's Orange Pippin and Lane's Prince Albert on new selected rootstocks. Averages per tree.

Reference No.	Parentage: Malling Rootstocks Nos.	Budded with Cox's Orange Pippin.				Budded with Lane's Prince Albert.					
		No. of trees.	Wood growth 3 years' total cm.	Fruit buds.		No. of trees	Wood growth 3 years' total. cm.	Fruit buds.		Fruits.	
				3 years' total	No. per metre of wood.			3 years' total.	No. per metre of wood.	Total No.	Number to the pound.
3426	VII×IX	29	133	5·1	3·6	—	—	—	—	—	—
346	IX×I	38	155	4·5	2·6	6	750	103	11·5	4·0	3·3
3412	II×IX	20	155	2·8	1·7	4	501	86	13·1	3·9	6·4
3411	IX×I	46	158	1·7	1·1	10	884	95	10·1	3·0	4·2
3418	IX×IV	39	163	4·8	2·9	6	448	46	10·2	0·9	3·3
3424	IX×V	43	174	12·9	6·9	9	315	54	13·7	1·5	3·6
3413	II×IX	39	181	10·8	5·8	12	645	136	16·4	6·0	4·0
343	I×IX	14	185	1·1	0·6	4	1,064	85	7·8	5·6	5·6
341	I×IX	41	188	2·9	1·5	10	855	128	14·2	0·3	2·9
3419	IX×IV	32	189	10·3	5·4	4	473	79	16·2	3·0	2·9
3415	II×IX	7	194	3·8	0·5	6	243	85	23·6	1·8	5·6
3431	XIII×IX	25	204	12·8	5·5	10	407	116	18·6	3·9	4·2
3427	VII×IX	48	207	1·0	0·5	8	726	110	12·4	5·8	3·3
3425	VI×IX	22	207	0·1	0·1	7	682	83	10·9	3·2	3·0
IX		**235**	**209**	**10·9**	**5·1**	**17**	**816**	**164**	**16·0**	**9·8**	**3·3**
347	IX×I	38	211	1·2	0·6	15	1,037	131	12·2	2·1	3·3
3410	IX×I	36	217	5·3	2·4	15	1,045	120	11·1	4·7	3·3
3416	IV×IX	31	217	7·0	3·2	9	429	67	12·4	1·2	4·8
3420	V×IX	41	219	4·2	1·9	5	581	96	10·7	2·8	3·9
3436	XVI×IX	37	230	3·4	1·4	9	1,377	330	18·9	14·0	3·0
3422	V×IX	46	245	1·0	0·4	29	1,322	249	13·4	10·7	3·1
3423	IX×V	46	259	4·3	1·0	28	880	179	12·8	12·7	3·8
II		**20**	**262**	**3·5**	**0·9**	—	—	—	—	—	—
342	I×IX	14	268	0·0	0·0	6	1,252	110	8·7	4·1	3·4
417	IX×IV	22	272	1·8	0·7	18	550	58	10·1	2·3	3·6
434	XV×IX	20	273	1·1	0·4	11	835	114	11·4	4·0	3·0
348	IX×I	19	274	1·7	0·6	7	1,111	70	6·3	4·0	3·6
433	XV×IX	20	283	4·8	1·5	11	842	145	14·3	5·7	3·4
428	IX×VII	43	284	7·0	2·5	6	1,795	208	10·5	8·5	3·0
344	I×IX	36	289	0·2	0·1	8	997	128	12·7	6·5	3·4
432	IX×XIII	19	290	0·5	0·1	9	710	99	13·0	7·6	3·1
421	V×IX	34	293	5·0	1·4	10	466	118	15·8	3·6	3·7
349	IX×I	41	296	0·5	0·2	8	868	56	6·3	3·9	3·3
437	V×IX	44	324	5·0	1·3	20	1,416	288	14·8	20·2	3·3
438	IX×XII	27	326	0·0	0·0	7	2,158	220	9·9	10·7	3·0
429	IX×VII	50	326	1·6	0·5	10	1,511	167	9·9	8·5	3·2
345	I×IX	31	336	0·4	0·1	7	1,255	138	10·9	6·5	3·2
414	II×IX	40	354	4·5	1·3	7	938	117	11·4	2·9	3·9
XVI		**44**	**370**	**0·0**	**0·0**	—	—	—	—	—	—
435	IX×XV	46	408	9·9	2·0	17	1,043	202	16·4	5·4	2·3
XII		**39**	**496**	**0·0**	**0·0**	**12**	**967**	**10**	**1·1**	**0·6**	**3·5**
XIII		**41**	**577**	**0·0**	**0·0**	—	—	—	—	—	—
430	XII×IX	36	874	0·3	0·03	14	1,519	171	10·4	4·5	3·6

In judging the significance of differences between rootstocks the following rough rule may be used for the wood growth with Cox's Orange Pippin:

Two rootstocks differ significantly ($P = 0{\cdot}05$) if the wood growth of one exceeds the wood growth of the other by 25%, and if both are represented by 20 or more trees.

This rule was obtained by examination of the variation within rootstocks 3426, 341, M.IX, M.II, 3428, 3429 and 3430.

growth had been found more suitable than Lane's Prince Albert for the relatively closely planted, intensive nursery trial contemplated, and it had also become a much more important commercial variety. The original scheme had been to replant the trees as maidens in a carefully designed field trial, but the outbreak of war made it necessary to utilize the land for more urgent purposes and the trees remained *in situ* at about 1½ feet apart in rows 4 feet apart until the end of 1942. They received routine spraying and cultural treatments but were not pruned. As far as practicable full records were kept on these trees throughout.

The records of wood growth, blossom formation and fruit production on the trees on Lane's Prince Albert and Cox's Orange Pippin are summarized in Table I and illustrated diagramatically in Fig. 1.

Rootstock effect on vigour.

Both Cox and Lane's proved quite compatible with all the thirty-eight selected rootstocks in this trial. The take of the buds of Cox was as high as 96 per cent. with two rootstocks and

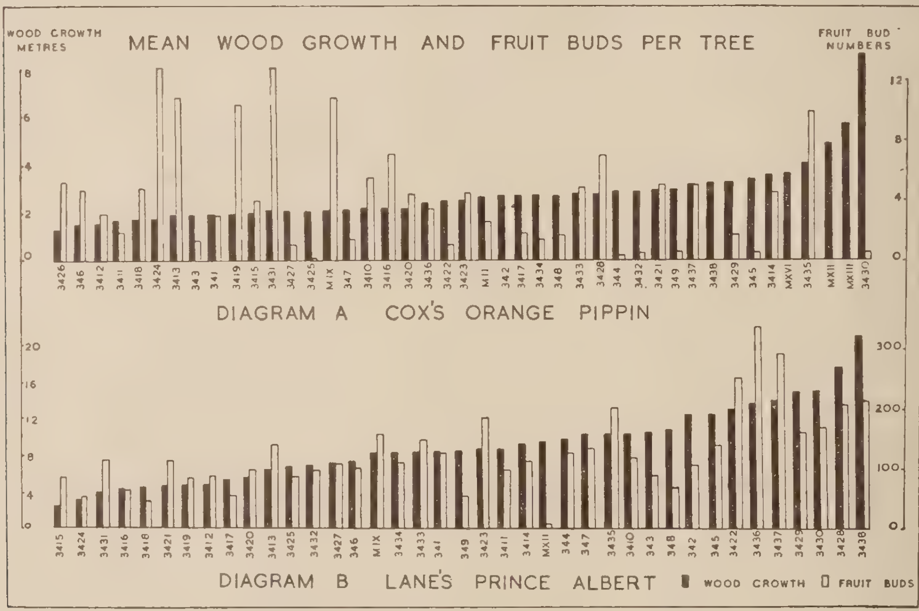

Fig. 1

Diagrams showing wood growth and fruit buds per tree.

fell below 50 per cent. in only two instances. No subsequent losses occurred which could be attributed to incompatibility between rootstock and scion.

From the totals of new wood growth shown in Table I, it is clear that these families have given rootstocks showing an extremely wide range of variation for vigour. With Cox's trees on 3430, the most vigorous rootstock, produced more than six times the wood produced by 3426, the most dwarfing one. With Lane's, the range was even greater, trees on the strongest rootstock having nine times the growth of those on the weakest one. When worked with Cox's, some of the rootstocks, produced significantly less total wood growth than Malling No. IX, while one gave a significantly larger total than trees on No. XII. Worked with Lane's, fifteen of the rootstocks were more dwarfing than No. IX and fourteen more vigorous than No. XII; but in this trial the trees on No. XII were unquestionably much weaker than is normal, possibly due to their occupying a rather unfavourable situation. The extreme range of difference in

wood growth between the strongest and weakest of these rootstocks, where worked with Lane's, was very much greater than that found among the nineteen seedlings from No. VIII × No. IX studied previously.

In the Table no attempt has been made to group the thirty-eight rootstocks according to vigour, based on records of wood growth; indeed, with both varieties they appear to form a continuous series from the most dwarfing to the most vigorous. By reference to the trees on the five Malling rootstocks (included for comparison) which represent Hatton's original grouping, it would appear that this new series extends much beyond the range of the Malling series, not only in the direction of much greater dwarfing but also in that of greater vigour. Genetically the more dwarfing rootstocks would appear analogous to the extremely dwarfing varieties already reported as occurring among seedlings of No. VIII × No. IX.

Probably the most notable single rootstock in the whole series from the point of view of vigour is 3430 which, with Cox's, made almost twice as much wood growth as the very vigorous No. XII in the three seasons. As already stated, the behaviour of No. XII was not typical in the series worked with Lane's, and comparison of the new rootstocks with standard types is not easy; but it seems fairly clear that several of these rootstocks made very vigorous trees while a number were outstandingly dwarfing.

Correspondence between the rootstocks where worked with Cox's and with Lane's, while generally fairly close, was not by any means complete. While this may be partly due to variations inherent in all such experimental work, it has already been established in studying the Malling series that different scion varieties may respond in different ways to the influence of a given rootstock. The most outstanding instances of this were 343, 347 and 3410, all of which were relatively dwarfing with Cox's and vigorous with Lane's, while with 3421 exactly the reverse was the case. Numerous less striking instances of the kind can be seen by reference to Table I; but, speaking very generally, correspondence between the two sets of trees was, on the average, fairly satisfactory.

ROOTSTOCK EFFECT ON TREE ANCHORAGE AND FLEXIBILITY.

The effectiveness with which the root system is able to anchor the tree in the ground is a well marked rootstock characteristic. It is a common criticism of Malling No. IX that it affords a poor anchorage, and the fact that young trees on this rootstock have to be staked adds greatly to the expense of establishing plantations.

The trees of Lane's on these thirty-eight selected rootstocks were all secured to wires and no differences in anchorage were observable; but, in reviewing the nursery of Cox's trees on the same stocks, which had remained unsupported until the end of their third year, it was apparent that there were very large differences in anchorage. On certain rootstocks the trees were lying almost prone while adjacent sets of trees on others were quite erect and securely held. The results of observations of this character, following a period of high wind in the autumn of 1942, are given in Table II.

On only three of these rootstocks were the trees recorded as poorly anchored and they were all of dwarfing or semi-dwarfing influence. Of the trees on rootstocks of approximately similar scion influence to No. IX, trees on four were well anchored and on none were the trees as poorly anchored as those on No. IX. Indeed, to those who think that a dwarfing rootstock must inevitably be poorly anchored it may come as something of a surprise to learn that many of these extremely dwarfing rootstocks were as firmly anchored as Nos. XII or XVI. The trees on the more vigorous of these new rootstocks were, with very few exceptions, all well anchored. The differences in anchorage found among these rootstocks were unquestionably the reflection of variations in the conformation of the root systems. The morphology of these root systems has been studied on the three year-old trees and will form the subject of a later communication.

Apple varieties differ very greatly in the resistance which their shoots offer to bending. On certain varieties the shoots can be bent into a complete ring without breaking, on others

they are so brittle that very little curvature is sufficient to snap them. Differences in flexibility, almost as great, may occur among trees of a single variety when worked on different rootstocks. Observations on the plantation of Cox's on these new rootstocks showed that on certain of them the trees, although well anchored, were unable to remain completely erect due to the flexibility of the wood. The results of these observations are shown in Table II. Actually some of the

TABLE II.

Relative tree anchorage and flexibility of shoots. Cox's Orange Pippin on new selected rootstocks.

Rootstock.	Anchorage.	Flexibility.	Rootstock.	Anchorage.	Flexibility.
3426	3	3	3423	3	1
346	2	1	II	1	1
3412	2	2	342	1	1
3411	1	1	3417	1	2
3418	1	1	3434	1	2
3424	2	2	348	1	1
3413	2	2	3433	1	2
343	1	2	3428	1	2
341	1	1	344	1	2
3419	2	1	3432	1	1
3415	3	2	3421	2	1
3431	2	2	349	1	1
3427	1	1	3437	2	2
3425	1	2	3438	1	2
IX	3	3	3429	1	1
347	1	1	345	1	2
3410	1	1	3414	1	1
3416	2	1	XVI	1	1
3420	2	1	3435	2	2
3436	1	2	XII	1	1
3422	1	1	XIII	1	1
			3430	1	1

Description of categories : Anchorage, 1 Good, 2 Fair, 3 Poor ; *Flexibility,* 1 Very stiff, 2 Fairly stiff, 3 Flexible.

worst affected trees were on No. IX and Seedling 3426, where a combination of poor anchorage and extreme flexibility made the trees very difficult to manage without support. Even among the more vigorous rootstocks with good anchorage, however, the trees were often unable to remain completely upright because the stems were not stiff enough to support their weight.

ROOTSTOCK EFFECT ON BLOSSOM PRODUCTION.

In Table I are shown the average number of blossom trusses borne by the trees of Cox's and Lane's during their first three years. As happens invariably the trees of Lane's were much more prolific at this early stage than those of Cox's. This was particularly marked in the more vigorous rootstocks. As an extreme example Seedling 3438 among the vigorous rootstocks had no blossom with Cox's but was one of the most prolific of the whole series with Lane's.

Although it is not very clear from the accumulated totals of the three years, given in the Table, there was, in general, the usual inverse relation between the vigour and the precocity of these rootstocks. With the more slowly maturing Cox's, the averages of total blossom trusses formed by trees on the various rootstocks show a fairly consistent decline from the more dwarfing to the most vigorous. The trees on the extremely dwarfing rootstocks appear to form an exception, but this is probably due to their very diminutive size.

With Lane's, the stage at which mere tree size directly influenced total blossom production was reached at a much earlier period and there was a direct positive relation between wood growth and number of blossoms by the end of the third year. That these results were merely

the consequence of the much greater amount of wood which trees on the vigorous stocks provide for the production of flowers is indicated by the totals of blossom trusses borne per metre of wood, also shown in Table I. With both Cox's and Lane's, if the trees on the extremely dwarfing rootstocks be excepted, there is a fairly regular decline in the number of flowers borne per unit of wood, with increasing vigour of the rootstock.

Rootstock effect on fruit production.

The trees of Lane's on these new rootstocks were subjected annually to severe attacks of Apple Blossom Weevil, while the fruiting of the Cox's may have been influenced to some extent by the fact that varieties suitable for cross pollination were not distributed evenly throughout the trial. In spite of this there was close correspondence between blossom formed and fruits matured with both Lane's and Cox's. Only the results for Lane's have been thought worth including in Table I. With Lane's, trees on a number of the more vigorous rootstocks had produced more fruit than those on the very prolific No. IX by the end of the three years ; but with Cox's the number of fruits borne by the trees on No. IX was exceeded only by those on trees on the very dwarfing Seedling 3413.

As an indication of the relative size of the fruits on different rootstocks the average number of fruits per pound was calculated for the Lane's and is shown in Table I. With both Cox's and Lane's the fruits from trees on the more dwarfing rootstocks were larger than those from trees on the more vigorous rootstocks, and there was a gradual decrease in size of fruit with increase in size of tree. It has already been well established that fruit from trees on Malling No. IX is larger on the average than that from trees on the more vigorous of the Malling series.

Slight differences in colour and finish were also noted on the Cox's. The fruits from trees on certain rootstocks were much brighter in appearance than those from others. These differences, although apparent to the eye while the fruits were on the tree, were not sufficiently definite to be significant when the fruits were compared in detail.

Rootstock effect on some physiological characters.

In general, the trees in both these trials were very free from any pathological troubles. Apart from the severe annual attacks of Apple Blossom Weevil on the Lane's already alluded to, which the situation of the field in which they were growing made it difficult to control, the routine sprayings were effective in keeping the more common diseases and pests in check on both Cox's and Lane's. A certain amount of Scab occurred on the fruits of Cox's in the only season in which they bore any considerable crop, but it was fairly general and the differences from rootstock to rootstock were not large enough to be significant.

There were, however, a number of troubles noted on the Cox's trees which were probably more physiological than pathological in their origin and over which the rootstock did appear to exert an influence. A relatively early season of leaf-drop has always been a characteristic of Cox's and, on these rootstocks, considerable differences were noted in the time at which the annual defoliation occurred. In Table III are shown the results of observations made on September 22nd, 1942. The percentage defoliation on the leaders, laterals and spurs of trees on the various rootstocks was noted.

There were two rootstocks the trees on which had retained all their leaves in late September, the dwarfing Seedling 3415 and the vigorous Malling No. XVI. Trees on both No. IX and No. II were severely defoliated, while those on No. XII had lost a considerable proportion of their leaves. It should be remembered, however, that the trees were growing extremely close together on land which was probably in rather poor condition. There seems to be little evidence to show that trees on the more dwarfing rootstocks in general lost their leaves at an earlier stage than those on the more vigorous ones, but there appears to have been an exceptionally severe early defoliation of the spurs on trees on some of the very dwarfing rootstocks.

To what extent foliage retained on the tree by the latter part of September is contributing anything to its nutrition is not precisely known. Nor is it known how far the early severe defoliation of these young Cox's on certain rootstocks was a normal process in a healthy tree, or how far it was a reflection of abnormal conditions. But the differences found between trees on different rootstocks, varying from an 85 per cent. defoliation of the leaders to none at all, on the same date, afford a striking instance of the profound effect which the rootstock can exert on the physiology of the tree.

The Cox's trees in these trials have always shown some symptoms of leaf scorch. Records were kept season by season and the results of a survey made at the end of the third season are summarized in Table III. Two types of scorching were distinguished ; marginal scorch, which

TABLE III.

Percentage of leaves lost on 22.9.42 and relative incidence of marginal and interveinal leaf scorch. Cox's Orange Pippin on new selected rootstocks.

Root-stock.	Defoliation. % of leaves lost.		Leaf Scorch.		Root-stock.	Defoliation. % of leaves lost.		Leaf Scorch.	
	Leader and laterals.	Spurs.	Marginal.	Interveinal.		Leader and laterals.	Spurs.	Marginal.	Interveinal.
3426	75	40	0	3	3423	50	30	0	1
346	65	0	2	0	II	35	0	2	0
3412	55	0	2	1	342	40	0	0	0
3411	80	40	0	3	3417	65	0	0	1
3418	70	30	0	3	3434	50	5	1	1
3424	60	10	1	2	348	70	0	1	1
3413	45	0	1	0	3433	70	20	0	1
343	60	10	0	1	3428	65	10	1	1
341	85	10	1	1	344	55	0	2	1
3419	85	30	1	2	3432	40	0	0	1
3415	0	0	1	0	3421	55	0	1	1
3431	45	20	1	1	349	55	10	1	1
3427	75	20	2	2	3437	40	10	1	0
3425	40	10	0	1	3438	40	0	1	1
IX	50	10	0	1	3429	45	10	1	0
347	55	0	2	0	345	80	0	1	1
3410	50	10	1	1	3414	30	0	2	0
3416	80	30	1	2	XVI	0	0	0	0
3420	60	0	2	0	3435	20	0	1	0
3436	40	0	0	0	XII	25	0	1	0
3422	75	10	2	1	XIII	5	0	1	0
					3430	10	0	0	0

Key to categories for leaf scorch : 0 None, 1 Little, 2 Moderately severe, 3 Very severe.

appears as a narrow band of brown tissue round the edges of the leaves and is usually regarded as a symptom of potash deficiency, and interveinal scorch, which commonly occurs in the form of brown discoloured areas in the middle portions of the leaf, and is attributed to lack of magnesium.

There were no instances of really severe marginal leaf scorch on trees on any of these rootstocks. Trees on No. II and on eight of the new rootstocks were moderately affected, while trees on fourteen rootstocks, including No. IX, No. XVI and the extremely vigorous 3430, showed no symptoms. This type of scorching occurred on trees on rootstocks of all degrees of vigour and there was no evidence that it occurred with greater intensity on trees on the more dwarfing or the more vigorous rootstocks.

With interveinal scorch, on the other hand, the only really severe cases recorded were on trees on the extremely dwarfing rootstocks. It is interesting to notice here the apparently close correlation between severe interveinal scorch and early defoliation of the spurs. Trees on all three rootstocks which had much interveinal leaf scorch all had exceptionally severe early defoliation of the spurs.

DISCUSSION.

Genetical aspects.

Progress in the elucidation of the genetical inter-relationships of apple varieties has been made difficult by the complex nature of inheritance in the Pomoidae. Cytologists have found a sufficient explanation for these complexities in the secondary polyploid condition of the nucleus which they are able to infer from chromosome behaviour in the species. Thus the difficulties of interpreting genetical results with characters which express themselves directly are sufficiently great, but they are considerably magnified when, as in rootstocks, important genetic characters must express themselves through a second variety, the scion, of different genetic constitution. Here the interaction of two dissimilar genetic systems must be considered, and results arrived at from comparisons of rootstock differences with the same scion variety may have to be extensively modified when another scion variety is used.

In spite of this it has been possible to distinguish, in a general way, certain well defined principles of inheritance operating in these crosses between the so-called Paradise apples, for which the existence of cumulative or additive factors may afford the best explanation. Crosses between the two most dwarfing rootstocks of the Malling series gave very vigorous rootstocks as well as some very considerably more dwarfing than either parent. With the present series, a seedling of No. IX crossed with the vigorous No. XIII was only about as vigorous as No. IX with both Cox's and Lane's, while the most vigorous rootstock in the whole series, 3430, was significantly more vigorous than its parent, the very vigorous No. XII. Seedlings from No. IX × No. II made rootstocks not only much more dwarfing than either parent but also very considerably more vigorous than the fairly vigorous No. II.

With respect to vigour in crosses between these rootstocks, the progeny does not by any means always approximate to an average condition between the parents, but may go much beyond the expected range in the direction of both greater vigour and more extreme dwarfness. The factors involved cannot therefore be single or simple and are most probably re-duplications of the type already postulated to explain the inheritance of other characters in apples.

On early fruitfulness in these rootstocks the influence of the precocious No. IX appears to have been more direct. The close association between a dwarfing influence on the scion and a tendency to produce early heavy crops, so marked in the Malling series, is still clearly shown, although there were occasional rootstocks, such as 343, 347, 3425 and 3427, which were dwarfing in their influence on the scion but not fruitful in the early years, particularly with Cox's. In these families of seedlings it does seem as though a certain measure of precocity has been inherited from the early bearing No. IX, even by the more vigorous rootstocks. Whereas the vigorous Nos. XII, XIII and XVI bore neither flowers nor fruits in the three years with Cox's, seedlings, of approximately equal or greater vigour, produced fair quantities of both. Of these seedlings 3435 was outstanding. More vigorous than No. XVI and only very slightly less vigorous than No. XII, trees of Cox's on it bore almost as many flowers and almost twice as many fruits as those on No. IX. Similarly with Lane's, trees on 3428, 3437, 3438 and 3430 showed an almost equally striking combination of early vigour and fruitfulness. Thus early vigour may be found in combination with early fruitfulness, at any rate in exceptional cases.

It is not easy to trace the direct influence of the parents in many of the other characters studied. No. VII crossed with No. IX gave Seedling 3426, with poor root anchorage and flexible shoots, as well as 3427 with good anchorage and stiff shoots. The same family that produced 3415, trees on which had no early defoliation whatever, also gave 3412, the

trees on which had lost 55 per cent. of the leaves from their leaders by late September. Instances of this kind could be multiplied, all illustrating how difficult it would be to attempt to predict the results to be expected from any particular cross. With leaf scorch there was some evidence to show that trees on rootstocks with No. II or V parentage were specially subject to potash deficiency marginal leaf scorch, and that those with Nos. I or IV parentage were more severely affected by the magnesium deficiency type than others. But even here there were some exceptions.

Practical aspects.

When it is remembered that the sixteen varieties of so-called Paradise rootstocks, now universally known as the Malling series, were most of them survivors of the old-fashioned fruit or kitchen garden, often selected by chance and casually introduced, it may be thought remarkable that they have adapted themselves as well as they have to the requirements of modern competitive commercial fruit growing. Yet under this much more stringent test many of these rootstocks have been found unsuitable and have been discarded, while, in those which remain, it has not been difficult to discover outstanding defects. No. IX, excellent in many respects and on many soils as a small, quick cropping filler, will rarely support itself satisfactorily. Of the semi-dwarfs, No. II is not quite the ideal rootstock for the nurseryman, while No. V has been all but completely discarded owing to its poor cropping and susceptibility to leaf scorch. Trees on No. XII are generally planted as vigorous standards, but there are many growers who are not unnaturally impatient at their tardy cropping. A better anchored No. IX, a more easily rooted No. II, or an earlier cropping No. XII, to mention only a few, would all save many thousands of pounds in the aggregate to the apple growing industry.

It was with these facts in mind that systematic breeding work was started at East Malling. The very serious losses incurred through the attacks of Woolly Aphis, particularly in the Dominions, made the production of Woolly Aphis-resistant rootstocks a matter of importance, and the considerable progress made in this work has been described elsewhere (Crane and others, 1936 ; Beakbane and others, 1941). Resistance to insect attack, however, remained only one of the objects which it was sought to attain. While the combination, in a series of rootstocks, of resistance to insect and disease attack with the ideal pomological characteristics already enumerated must be the ultimate goal, it was realized that this could be attained only by the gradual accumulation of small improvements. It was logical, therefore, that side by side with the attempt to breed resistance to Woolly Aphis into the existing rootstocks there should also be an attempt to improve their pomological performance. The first object could be realized only by bringing in resistance from varieties outside, the second might be achieved by extensive interbreeding.

There seem some grounds for the expectation that several of these new rootstocks may prove, on further trial, to be decided improvements on those at present in use. The extremely vigorous 3430, while it has made larger trees than No. XII, has also shown signs of being more precocious. In 3435 the qualities of early vigour and early cropping seem to have been combined to quite an unusual degree. Among the rootstocks similar to No. II in their influence on the scion, there are several somewhat more ready rooting and without the susceptibility to leaf scorch so characteristic of Nos. V and II. None of the rootstocks of approximately similar vigour to No. IX was as poorly anchored, while several had an outstandingly good root-hold and withstood the effects of several severe gales. For the extremely dwarfing rootstocks it is not so easy to foresee a future. But for very intensive work, on soils which induce very vigorous growth, stocks of this type may be found to have their uses.

A selection of what are at present considered to be the best of these thirty-eight rootstocks, largely based on their performance with Cox's, has been made, and the three-year-old trees of Cox's on them have been planted out in a carefully designed field trial. The selected stocks are :

Vigorous.	*Intermediate.*	*Dwarfing.*
3432 (No. IX × No. XIII).	3420 (No. V × No. IX.)	3426 (No. VII × No. IX.)
3437 (No. V × No. IX).	3436 (No. XVI × No. IX).	3424 (No. IX × No. V).
3438 (No. IX × No. XII).	3422 (No. V × No. IX).	3413 (No. II × No. IX).
3414 (No. II × No. IX).	3423 (No. IX × No. V).	341 (No. I × No. IX).
3435 (No. IX × No. XV).	3421 (No. V × No. IX).	3419 (No. IX × No. IV).
3430 (No. XII × No. IX).	3428 (No. IX × No. VII).	3431 (No. XIII × No. IX).

Observations on these trees will be continued for a further period with a view to a final rigid selection and introduction.

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SUMMARY.

Details of the behaviour during three years of Cox's Orange Pippin and Lane's Prince Albert apples worked on thirty-eight rootstocks selected from families of crosses between the Malling series of Paradise are given.

All the rootstocks proved compatible with Cox's and Lane's, but there were wide variations in the percentage bud take with Cox's.

They showed very great differences in their capacity to induce vigour in both scion varieties, the most vigorous making trees considerably stronger than those on No. XII, and the more dwarfing, trees much smaller than those on No. IX.

While, in general, there was the usual very close relation between the dwarfing character and early productivity, several of the more vigorous of these new rootstocks gave evidence of being much more precocious than the vigorous rootstocks of the Malling series.

Trees on these rootstocks were found to differ greatly with respect to such characters as anchorage, relative flexibility of shoots, time of autumn defoliation and susceptibility to both marginal and interveinal leaf scorch.

The genetical implications of the results are discussed, and it is suggested that such characters as vigour in these rootstocks are controlled by reduplicated factors which are cumulative in their effects.

It is suggested that certain of these rootstocks may be found in practice to be an improvement on those at present in use in certain definite respects. The lines along which future work will proceed are indicated.

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STUDIES ON THE VEGETATIVE PROPAGATION OF FRUIT TREE ROOTSTOCKS

II. BY HARDWOOD CUTTINGS

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INTRODUCTION.

Although certain fruit tree rootstocks, such as those of the Marianna plum, can be propagated successfully by hardwood cuttings, others, such as the Pershore plum and the various apple rootstocks, cannot be multiplied satisfactorily in this way owing to the failure of a large proportion of them to develop roots. The conditions governing root production in cuttings have been studied by many investigators and a review of their work has recently been undertaken by Garner (1943). The effect of growth substances (auxins) has received special attention recently and the work on this subject has been discussed by Thomas (1937), and by Pearse (1939). Further studies, particularly with those rootstocks which would be commercially valuable if they could readily be propagated vegetatively, seemed eminently desirable, and the present paper deals with the methods and results of the experimental work done on the problem at East Malling in 1939 and 1940.

METHODS.

In order that the results of the trials might be subjected to statistical analysis, the beds of cuttings were laid out in blocks, randomized plots and sub-plots. The unit was one row, consisting of 12 cuttings in the first experiment, 20 in the second, and 10 in the third. The cuttings, retaining their upper leaves, were taken in autumn, planted in soil in the first experiment and in sand in the other two. Treatment with auxins was by Hitchcock and Zimmerman's (1936) immersion method, the time being 24 hours, and the cuttings were rinsed in tap water immediately after treatment. At the end of the growing season records were taken of (i) the number of rooted cuttings per row (the most important index of success), (ii) the number of roots produced, (iii) the length of roots, and, usually, (iv) the length of shoot per cutting. Only such cuttings as were fully alive and possessed roots were counted as " rooted ". The number of cuttings per treatment, or combination of treatments, varied in the different experiments; but for the sake of uniformity the numbers have been expressed as percentages in all the Tables. The significance of differences between the values for different treatments and combinations of treatment, was determined by analysis of variance. As the percentages were based on small whole numbers, angular transformation of them into Bliss's variate was carried out in accordance with the recommendations of Bliss (1937 and 1938) and Cochran (1938). The variate values were taken from Fisher and Yates's (1938) Table XIII ; the other data were analysed untransformed.

EXPERIMENTAL.

Experiment I.

This was carried out to investigate the effect of (*a*) source of material, (*b*) position of the apical cut, and (*c*) irrigation, on the root and shoot growth of hardwood cuttings of the plum rootstock Myrobolan B. The cuttings, from one-year-old shoots, were taken in October from

* These studies were carried out by the first author during a period spent at the East Malling Research Station, Kent, England, and formed part of a Thesis approved for the Degree of Doctor of Philosophy in the University of London. Part I was published in this Journal, Vol. XX, 1942, pp. 1-11. The present paper has been edited and adapted from part of the thesis by the second author, under whose direction the studies were carried out, and who accepts responsibility for any modifications involved.

(i) a hedge, (ii) a layer-bed, (iii) budded layers, (iv) one-year-old rooted cuttings, and were planted at once in the normal soil of an experimental plot. The hedge was a well established one on its own roots, and the shoots used arose from a mature scaffolding of branches. The layer-bed (ii) was likewise well established, but had received the equivalent of severe stem pruning every year ; the shoots used arose directly from the buried layers. The budded layers (iii) were the rooted shoots removed from a layer-bed the previous autumn and planted out about one foot apart in nursery rows, with such roots as they then possessed. They had thus received the equivalent of severe root pruning. They had produced lateral shoots, and it was from these that the cuttings were taken. The one-year-old rooted cuttings (iv) had been taken from the main shoots on a layer-bed the previous autumn and possessed no roots when planted. The cuttings used were taken from the lateral shoots which had since developed.

In making the cuttings, the upper cut was made either just above or just below a bud, so that either only a short length of internode or a whole internode was left above the apical surviving bud.

Two treatments were tried : (i) non-irrigated or dry, as a control, and (ii) irrigated. A moisture meter was installed in each plot, the centre of the pot being 5 in. below the surface of the soil and about 1 in. from a row of cuttings. The meters were set at zero at the start and the pots were refilled, and the meters reset at zero, twice during the season. A known amount of water was applied to the irrigated plots whenever the moisture tension in them rose to 20 cm. The tensions, as cm. mercury, the daily rainfall and the amounts of water given to the irrigated plots were recorded but are not given here. Up to the time of the first irrigation, the readings on both meters were about the same, later the tension rose in the drier non-irrigated plot. This drying was confirmed by soil moisture determinations carried out on samples taken at three different depths from the two plots. The total rainfall was comparatively high, however, during the critical period for growth in 1939, and the soil even in the non-irrigated plots never got very dry.

The experiment was laid out in three blocks of four randomized plots, one for each combination of position of top cut and irrigation treatment. These plots were divided into four sub-plots, arranged at random, one for each source of cutting. Each sub-plot consisted of five rows of twelve cuttings.

The weight and mean diameter (from 50 cuttings), and the mean number of nodes per cutting (from 100 cuttings) were determined from samples from each before planting, and are shown in Table I, from which it is seen that the cuttings from budded layers were slightly heavier, had a greater mean diameter and shorter internodes than the others.

TABLE I.

Weight, mean diameter and number of nodes at time of planting. Myrobolan B.

Source of cutting.	Weight of 50 cuttings (oz.).	Mean diameter in mm.	Mean number of nodes.
Hedge	2·0	2·74	21·9
Layer bed	2·0	2·68	21·6
Budded layers	2·3	3·12	24·5
1-year cuttings	2·0	2·80	21·8
Significant Difference ..		1·2	0·3

At the end of the growing season the number of rooted cuttings and the total shoot growth per row were determined in the field. Cuttings from representative plots were dug and certain further measurements, including number and spread of roots and weights of the cuttings, were made on them. These measurements, however, revealed little of additional interest and are not given here.

Results.—The percentages of the cuttings which rooted are given in Table II. In the early stages the cuttings on the irrigated plots seemed the more promising, they had more shoot growth and darker leaves than those on the dry plots, but at the end of the season there was no significant difference between the mean values for the percentage rooted, viz. 56·6 and 54·9 per cent. respectively, for the irrigated and non-irrigated plots.

TABLE II.

Percentage rooting of Myrobolan B cuttings. Effect of source, irrigation and position of apical cut.

Irrigation.	Apical cut in relation to top bud.	Sources of cuttings.				Mean.
		Hedge.	Layers.	Budded layers.	One-year-old cuttings.	
Irrigated ..	Above	76·7	66·7	46·1	29·4	54·7
,, ..	Below	83·9	73·9	48·3	27·8	58·5
Non-irrigated ..	Above	75·6	59·4	40·6	20·0	48·9
,, ..	Below	83·3	75·0	52·8	32·8	61·0
Irrigated		80·3	70·3	47·2	28·6	56·6
Non-irrigated		79·4	67·2	46·7	26·4	54·9
Above bud		76·1	63·1	43·3	24·7	51·8
Below bud		83·6	74·4	50·6	30·3	59·7
Source		79·9	68·8	46·9	27·5	55·8
		All differences significant.				

The position of the apical cut had no significant effect on percentage rooting.

The source of the cutting had a very marked and significant effect. Cuttings from the hedge did best, with a mean value of 80 per cent.; those from laterals on one-year-old cuttings were worst, with a mean value of 28 per cent. Cuttings from the layer-bed did better than those from laterals on budded layers, the mean values being 69 and 47 per cent. respectively.

The mean shoot growth per cutting is shown in Table III. Cuttings from budded layers

TABLE III.

Mean shoot growth in cm.

Irrigation.	Apical cut.	Source of cutting.				Mean.
		Hedge.	Layer.	Budded layers.	One-year-old cuttings.	
Irrigated ..	Above	127·7	124·7	152·7	143·0	137·0
,, ..	Below	111·3	109·7	116·7	156·0	123·4
Non-irrigated	Above	86·0	85·7	140·0	131·7	110·8
,, ..	Below	115·7	95·0	138·7	148·7	124·5
Irrigated		119·5	117·2	134·7	149·5	130·2
Non-irrigated		100·8	90·3	139·3	140·2	117·7
Above bud		106·8	105·2	146·3	137·3	123·9
Below bud		113·5	102·3	127·7	152·3	124·0
Sig. Diff.						18·0
Source		110·2	103·8	137·0	144·8	124·9
		Sig. Diff.		14·5		

and from one-year-old rooted cuttings made significantly more growth than those from the hedge or the layer-bed. There was no significant difference between the values for hedge and layer-bed, or between those for budded layers and one-year-old cuttings. It is probable that the larger size of the cuttings from the budded layers and one-year-old cuttings was a spacing effect.

Experiment II.

This was planned to study the effects of etiolation, portion of shoot used and treatment with synthetic growth substances on rooting. Previous work by Vyvyan (1943) had shown for the plum rootstock Common Mussel, that etiolation of the base of the shoot improved the rooting of a basal etiolated cutting but reduced it in a cutting taken from the upper green part of the shoot. This suggested that etiolation might alter the distribution of some internal root producing substance, causing it to accumulate in the base. The experiment was repeated using more than one variety, and treatment with a growth substance was tried to see if this would serve as a substitute for etiolation.

TABLE IV.

Percentage rooting in Common Mussel and Pershore. Effect of etiolation of base of shoot, and of treatment with 20 p.p.m. indolebutyric acid on rooting of cuttings from tip, middle and base.

Auxin Treatment.	Treatment of base.	Common Mussel.				Pershore.			
		Tip.	Middle.	Base.	Mean.	Tip.	Middle.	Base.	Mean.
Untreated	Non-etiolated	(8·3	*16·7)	33·3	19·4	(2·5*	5·8	1·7)	3·3
	Etiolated	(7·6	*6·7)	58·3	24·2	(14·2	5·9	(10·0)	10·0
Treated	Non-etiolated	10·0	55·6	(77·5	47·7	6·7*	(20·0	20·8)	15·8
	Etiolated	(11·7	*17·5)	89·2	39·3	10·8*	(26·7	37·5)	25·0
Untreated (mean value)		(7·9	11·7)	45·9	21·8	(8·3	5·9	5·9)	6·7
Treated (mean value)		10·8	36·7	83·3	43·6	8·8	(23·3	29·2)	20·4
Non-etiolated (mean value)		9·2	36·3	55·4	33·6	4·6	(12·9	11·3	9·6
Etiolated (mean value)		(9·6	12·1)	73·8	31·8	12·5	16·3	23·8	17·5
Portion (general means)		9·4	24·2	64·4	32·7	8·6	(14·6	17·5)	13·5

Values in lines or columns enclosed in brackets, or distinguished by * are not significantly different.

Two varieties of plum rootstock, Common Mussel and Pershore, were used. In part of a layer-bed of each rootstock, the layers were pegged down and earthed over just before bud break, while in another part the layers were pegged down but not covered. Shoots arose from both sets of layers, but those from the earth-covered ones had etiolated bases, while the bases of the shoots from the other set were not etiolated.

At the end of the growing season, shoots were removed from both parts of the beds and three cuttings (10 in. long) were taken from each shoot, from tip, middle and base respectively. Where the shoots were longer than required to provide these three cuttings, the pieces between the tip and middle portion, and between the base and middle portion were discarded. It was noticed that the etiolated basal cuttings had fewer leaves than the non-etiolated ones.

The cuttings were subjected to two treatments, (i) distilled water, and (ii) indolebutyric acid in distilled water at a concentration of 20 p.p. million.

The experiment was laid out in six blocks, each with four plots arranged at random, one for each combination of the two varieties and two etiolation treatments. Each plot had six rows, arranged at random, one for each of the six combinations of portion of shoot and growth substance treatment. Each row had 20 cuttings.

Results.—The data for each variety were analysed separately. The mean values for percentages rooted are given in Table IV, and from the values the following points emerge :

(1) The values in line 3 show that in untreated and non-etiolated Common Mussel (C.M.), basal cuttings rooted significantly better than the others, and tip cuttings worse—but not significantly worse—than middle cuttings. In Pershore (P.), cuttings from all three portions did very badly and there were no significant differences, though there was some indication that basal cuttings were worst.

(2) The values in lines 3 and 4 show that, in Common Mussel, etiolation had no appreciable effect on tip cuttings but caused a great and significant increase in the rooting of basal cuttings. That of middle cuttings was decreased by etiolation, and although this reduction was not significant, the interaction between middle and basal portions, and non-etiolation and etiolation, was thoroughly significant. This result is in complete agreement with that found by Vyvyan (1943) for the same variety. Pershore behaved differently; etiolation improved rooting in both basal and tip cuttings, though the effect was significant only in the latter.

(3) Lines 3 and 5 show that in every case in both varieties cuttings treated with indolebutyric acid rooted better than the corresponding controls; except in tip cuttings, the differences were always significant and generally very considerable. This is not in agreement with the results found by most workers, who, in general, have obtained little improvement in the rooting of hardwood cuttings by treatment with this substance.

(4) The interaction between treatment and etiolation can be examined by comparing the sums of the values in lines 3 and 6 with the sums of those in lines 4 and 5. In no case was the difference between these summed values significant. In fact a comparison of the values in lines 5 and 6, with those in lines 3 and 4, shows that the effect of etiolation was much the same in each case. There were a few differences in degree ; for example, the reduction from 55·6 to 17·5 per cent. in the treated middle cuttings of Common Mussel is significant, whereas that from 16·7 to 6·7 per cent. in the corresponding untreated cuttings is not significant. Similarly, the increase from 77·5 to 89·2 per cent. in the treated basal cuttings of this variety is just not significant, whereas the corresponding increase from 33·3 to 58·3 per cent. in the untreated cuttings is significant. The interaction, however, between middle and base, and etiolation and non-etiolation, is as thoroughly significant in the treated cuttings as in the untreated.

From the values given in Table V it will be seen that neither treatment with indolebutyric acid nor etiolation of the base of the shoot had any marked or consistent effect on the mean length of a single root. Treatment, however, greatly increased the number of roots per cutting, especially those of middle and basal cuttings.

TABLE V.

Mean number of roots per cutting and mean length of single root.

Variety.	Treatment.	Etiolation.	Tip.		Middle.		Base.	
			Number	Length cm.	Number.	Length cm.	Number.	Length. cm.
CM	C	N	4·9	4·1	5·3	5·2	5·1	6·5
		E	3·4	4·2	3·6	5·8	7·2	5·5
	T	N	5·9	4·6	8·4	4·9	8·8	5·6
		E	7·4	4·4	8·4	4·9	10·4	5·4
P	C	N	6·0	3·3	3·7	4·4	2·0	6·8
		E	4·1	6·8	3·6	7·0	4·1	5·0
	T	N	8·6	6·6	10·8	5·5	11·7	4·3
		E	8·3	3·6	12·6	5·0	8·9	5·6

C = Control. T = Treated with growth substance. N = Non-etiolated. E = Etiolated.

Experiment III.

This was planned to investigate the effect of treatment, in light and in darkness, with various concentrations of indolebutyric acid and its potassium salt, on the rooting of the cuttings. The potassium salts of the auxins have been tried by several workers; Avery, Burkholder and Creighton (1937), using the Avena coleoptile test, found these salts even more effective than the acids. Grace (1937) got similar results. Zimmerman and Hitchcock (1937), using intact plants and cuttings, was unable to find much difference in effectiveness, but thought the potassium salt was less toxic at the upper limits of concentration. Later, however (1939), working with 70 species of plants, they found the salts better and attributed this in part to water solubility relations.

Two plum (Myrobolan B and Pershore), and two apple (Malling Nos. II and IX), rootstocks were used. Myrobolan B usually roots better than Pershore and No. II better than No. IX.

Indolebutyric acid and its potassium salt were each used at three concentrations, viz. 10, 20 and 40 parts per million. The controls received distilled water alone. Absorption of the solutions took place in (i) complete darkness, and (ii) continuous light from a 200 watt lamp 3 ft. away. The cuttings were planted in four blocks, each consisting of eight randomized plots—one plot for each combination of the four varieties and two light treatments. Each plot was made up of eight randomized rows—two control rows and one for each combination of the three concentrations and two substances.

Results.—The mean values for percentage rooting of the two plum rootstocks and the apple No. II are given in Table VI. The performance of the apple rootstock, No. IX, was too poor to be worth recording. In the statistical analysis, carried out after transformation of the percentages into Bliss's variate, the two plum varieties were combined, but the apple rootstock No. II was analysed separately. From the Table it will be seen that Myrobolan B rooted significantly better than Pershore. There is some indication that treatment in darkness gave better results than in light, but neither the effect of illumination nor its interaction with variety was statistically significant.

Statistical analysis showed that, taken as a whole, the differences between rows receiving unlike treatments were just significantly greater than those between rows receiving identical treatments. Treatment with either growth substance had clearly little or no effect, nor were there significant differences between the effects of the acid or salt, or the three concentrations. The only difference that was statistically significant was the second order interaction between substance, variety and light ; that is to say the relative response to the two substances in light and darkness respectively was different in the two varieties. Myrobolan B did best with the acid when treated in light and with the salt when treated in darkness. Pershore did best with the salt when treated in light and more or less the same with both substances when treated in darkness. Although this differential response was not very marked or very important, it gives some indication of the way in which alteration of even small details in method of treatment may have different results in different varieties.

Conclusions.

The results of the above experiments suggest that it would be unwise to draw general conclusions from observations carried out on single varieties. Not only do varieties differ widely in their rooting capacity, but they respond differently to identical treatments. The conditions best suited for rooting in one are not necessarily those for another.

For hardwood cuttings of Myrobolan B, source of material proved very important, and those taken from a hedge rooted best. It would be worth while examining other varieties to find out how far this applied to them. Special hedges might be planted, and cuttings from old trees, cordons and dwarf pyramids might be tried. With cuttings of this variety, planted in soil, neither irrigation nor the position of the apical cut had any effect ; but the season concerned was unusually wet and a different result might be obtained under drier conditions.

TABLE VI.

Percentage rooting in the plum rootstocks Myrobolan B and Pershore and the apple rootstock Malling No. II after treatment in light or darkness with various concentrations of indolebutyric acid or its potassium salt.

Number planted.		Myrobolan B.				Pershore.				Malling No. II.			
		Light.		Dark.		Light.		Dark.		Light.		Dark.	
320	Mean values.*	56·6		65·6		9·4		10·9		13·1		14·1	
		Acid.	Salt.	Acid.	Salt.	Acid.	Salt.	Acid.	Salt.	Acid.	Salt.	Acid.	Salt.
80	Control (2 rows per plot)	58·8		66·3		8·3		1·3		11·3		10·0	
40	10 p.p.m. (1 row per plot)	52·5	57·5	75·0	67·5	7·5	17·5	12·5	5·0	12·5	12·5	7·5	15·0
40	20 p.p.m. ,,	77·5	55·0	55·0	65·0	0·0	15·0	10·0	20·0	12·4	20·0	27·5	10·0
40	40 p.p.m. ,,	45·0	47·5	55·0	75·0	10·0	7·5	15·0	22·5	15·0	10·0	10·0	22·5
120	Substance (mean)	58·3	53·3	61·7	69·2	5·8	13·3	12·5	15·8	13·3	14·2	15·0	15·8
240	Treated (mean)	55·8		65·4		9·6		14·2		13·7		15·4	

* Based on four plots each made up of 8 rows of 10 cuttings.

Etiolation of the base of the shoot has in the past been regarded as beneficial, but the present experiments suggest that its effectiveness depends on the variety and on the portion of the shoot used. In the variety Pershore it seemed to improve the rooting of cuttings taken from all three parts of the shoot, but in Common Mussel the improvement was confined to basal cuttings that included the actual etiolated region ; rooting of cuttings taken higher up in the shoot was reduced.

Portion of shoot used proved important ; tip cuttings were of little use in the varieties tried. In Common Mussel, cuttings from the middle of the shoots gave good results only if the base of the shoots had not been etiolated.

Treatment with indolebutyric acid gave strikingly good results with two varieties of plum in one experiment, but in another, involving one of the same varieties, it caused no significant improvement. In this experiment, the acid and its potassium salt seemed equally ineffective, and absorption in complete darkness seemed slightly better than in continuous light ; but the difference was not significant.

SUMMARY.

Three experiments on the rooting of hardwood cuttings of fruit-tree rootstocks were carried out and the results analysed statistically.

1. Cuttings of the plum rootstock Myrobolan B were taken from four sources, with the top cut either just above or just below a bud, and planted in soil, either with or without irrigation.

(*a*) Source of material proved very important. Cuttings from a mature hedge rooted 80 per cent., from a layer bed 69 per cent., from budded layers 47 per cent., and from one-year-old rooted cuttings only 28 per cent.

(*b*) Neither position of top cut nor irrigation had a significant effect.

2. Cuttings of two plum rootstocks, Common Mussel and Pershore, were taken from the tips, middles and bases of shoots, whose bases either had, or had not, been etiolated. Some of each kind were treated with 20 p.p. million indolebutyric acid before planting, the remainder with distilled water.

(*a*) Common Mussel rooted much better than Pershore.

(*b*) Relative rooting of tips, middles and bases depended on variety and etiolation treatment.

(*c*) Previous etiolation of the base of shoot improved rooting in basal cuttings of both varieties and in tip cuttings of Pershore, but reduced rooting in middle cuttings of Common Mussel.

(*d*) Treatment with the growth substance greatly increased rooting in basal and middle cuttings of both varieties.

3. Cuttings of two plum rootstocks, Myrobolan B and Pershore, and two apple rootstocks, Malling Nos. II and IX, were treated, before planting, with either indolebutyric acid or its potassium salt, at three different concentrations, or with distilled water, either in continuous light or in darkness.

(*a*) Rooting in Myrobolan B was good, in Pershore and No. II moderate, and in No. IX too poor to be worth recording.

(*b*) Treatment had no significant effect, at any concentration, whether the acid or the salt was used, or whether carried out in the light or in darkness.

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STUDIES IN THE DIAGNOSIS OF MINERAL DEFICIENCY

I. THE DISTRIBUTION OF CERTAIN CATIONS IN APPLE FOLIAGE IN EARLY AUTUMN

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Leaf analysis has been very commonly used as a method for the diagnosis of mineral deficiency in fruit trees, as in other plants. Investigators have generally taken samples for analysis from leaves of one restricted type, on the very reasonable assumption that differences in age and position would be accompanied by differences in composition which might obscure the diagnosis. There has, however, been no systematic investigation of the mineral composition of different morphological types of leaf, or of the extent to which the different types reflect the nutritional level of the tree as a whole.

Most investigators have taken samples from the long shoots. Wallace (1940), for instance, normally takes samples from the base of these shoots, but where there is inadequate growth to provide a representative sample of this type—which often happens if the nutrition is unsatisfactory—leaves are taken from the non-fruiting spurs (Wallace, private communication). Roach (private communication), on the other hand, normally takes his samples from the youngest fully expanded leaves on the long shoots. Warne (1934), in an investigation of leaf scorch, sampled both apical and basal leaves on the terminal shoots and found the potassium content higher in the former. Reuther and Boynton (1940) sampled both shoot and spur leaves (corresponding in the notation to be used later in this paper to the middle L, and to the X samples) and generally found the potassium content of the latter to be lower. Larson (1933) sampled leaves from both fruiting and non-fruiting spurs; the latter contained more calcium, but there was little difference in the potassium content. Wallace and Mann (1926) report a single comparison between shoot and spur leaves—though the position of the leaves and the type of spur are not specified—in which the potassium and iron contents were much higher and the calcium and magnesium contents slightly higher in the shoot leaves. Wallace (1940) remarks that the leaves nearer the tip of the long shoots generally contain rather more magnesium than those at the base. Kidson *et al.* (1940) sampled leaves from the tips and lower parts of the leaders in an experiment on magnesium deficiency; they found the magnesium content higher at the tip, and the potassium content higher in the lower leaves, while the calcium contents were about the same. The range of magnesium content in trees showing various degrees of magnesium deficiency was greater in the lower leaves.

In view of the lack of information on the subject it was thought desirable to put in hand an investigation into the mineral content of different morphological types of leaf from apple trees growing under different nutritional conditions. The dessert apple manurial trial described by Hoblyn (1941) seemed to be a suitable source of material. The manurial treatments (begun in 1931) were originally 168 lbs. per acre of sulphate of ammonia, 224 lbs. sulphate of potash, and 560 lbs. superphosphate, applied annually in all combinations and replicated in eight blocks. Each plot receiving a different manurial treatment contained nine experimental trees, being certain combinations of two scion varieties with four clonal rootstocks. From 1937 onwards an additional 224 lbs. per acre of sulphate of potash was supplied annually to every plot, including those which previously had received no potash. In the spring of 1938, four out of every eight plots undergoing the same treatment received a dressing of humus, while the other four received over a period of three years additional fertilizer containing the same amounts of the mineral elements. On account of the large number of analyses involved it was necessary to limit to

sixteen the number of trees whose foliage was sampled ; samples were therefore taken only from one Cox's Orange Pippin tree on Malling rootstock XII (very vigorous) on each of sixteen plots. There appeared to be a nutritional difference between the two ends of the plantation, the trees at the east end looking much healthier, growing more strongly and cropping better than those at the more low-lying west end. The experiment was therefore arranged to include plots undergoing the eight manurial treatments (but none that had received the humus application in 1938) from both ends of the plantation.

The types of shoot included in the experiment were the primary long shoots, described by Vyvyan (1935) as type L, the primary non-fruiting spurs (Vyvyan's type V), and the secondary spurs from spurs bearing fruit (included in Vyvyan's type X). From each shoot three leaves were taken—that nearest the base, that nearest the apex, and one in the middle. Each sample consisted of two leaves—one from each of two shoots—and the samples were replicated four times from each tree. The samples were taken between September 17th and 19th, 1940.

The leaves were very carefully wiped with muslin in order to remove dust and spray residues, the laminae separated from the midribs and petioles and rapidly dried in a vacuum desiccator with silica gel. The lamina material was ground, and analysed by a spectrographic method of which details have not yet been published. In outline, the method was as follows : the material was folded into a spill of filter paper, carbonized in ammonium chloride vapour and fed mechanically into an oxy-hydrogen flame. A small amount of thallium chloride was pipetted on to each spill before carbonization to serve as an internal standard and to enable comparisons with spectra of a standard solution of metallic chlorides and phosphoric acid to be carried out. The elements for which data were obtained were calcium, iron, magnesium, manganese and potassium. It should be pointed out that, in view of the incipient stage of development of the method at the time when the analyses were carried out, too great reliance should not be placed upon the absolute values of the figures given.

Table I shows the mean mineral content for each tree. None of the differences in calcium, iron or magnesium contents is found to be significant if an error estimate is derived from the interaction between treatment and end of plantation. With manganese and potassium there is a highly significant difference between the trees at the two ends of the plantation, the leaves sampled at the west end containing only 75 per cent. of the potassium and 48 per cent. of the manganese found in those from the east end. Whether the low content of these elements at the west end of the trial is responsible for the poor performance of the trees at this end the data do not show ; it is, however, worth noting that there has been throughout the history of the trial a very strong response to potash application. The mean potassium content of leaves from the plots receiving the double application of potash is 58 per cent. higher than that found with

TABLE I.

Mean mineral content (dry matter basis) of all leaves from each tree sampled.

	End of plantation.	Treatment.							
		K	NK.	PK.	NPK.	2K.	N2K.	P2K.	NP2K.
Calcium (%) ..	East	0·71	0·53	0·42	0·54	0·59	0·38	0·60	0·60
	West	0·42	0·51	0·68	0·41	0·75	0·58	0·48	0·39
Iron (p.p.m.) ..	East	81	66	69	72	81	77	80	79
	West	68	75	70	61	77	61	77	74
Magnesium (%) ..	East	0·194	0·131	0·123	0·138	0·143	0·108	0·172	0·134
	West	0·125	0·154	0·180	0·097	0·194	0·161	0·171	0·103
Manganese (p.p.m.)	East	49	64	35	78	51	74	40	70
	West	22	28	25	20	32	39	27	28
Potassium (%) ..	East	0·51	0·74	0·55	0·76	0·93	1·20	0·86	1·00
	West	0·55	0·58	0·33	0·43	0·89	0·67	0·69	0·80

the single application. This difference is highly significant, as might be expected in view of the marked responses in growth and yield, and is greater than that between the two ends of the plot. Application of sulphate of ammonia has had the effect of increasing significantly (from 35 to 50 p.p.m.) the mean manganese content of the leaves. The increase in potassium content and the decrease in magnesium content with this treatment do not quite reach significance.

Table II shows the mean mineral contents of the various leaf types for all trees sampled. The effect of leaf position and its interaction with shoot type is significant for every element, and for all elements but iron the differences between shoot type means are also significant. With every element but potassium the highest content is found in the basal leaves of the non-fruiting spur, which are probably the oldest leaves sampled. There is a progressive decline in mineral content towards the apex of the non-fruiting spur; the secondary spur on a bearing spur generally has leaves with a still lower mineral content and showing little change from base to apex. In the long shoots the magnesium and calcium contents are highest in the basal leaves, and do not differ significantly between the middle and apical leaves; with iron the apical leaf

TABLE II.

Mean mineral content (dry matter basis) of different types of leaf.

Shoot Type.	Leaf position.	Calcium (%).	Iron (p.p.m.).	Magnesium (%).	Manganese (p.p.m.).	Potassium (%).
L	Apical	0·13	88	0·082	43	0·86
	Middle	0·20	70	0·058	33	0·97
	Basal	0·62	66	0·165	43	0·73
V	Apical	0·60	64	0·152	41	0·63
	Middle	0·85	88	0·204	51	0·63
	Basal	1·01	92	0·244	59	0·65
X	Apical	0·45	63	0·120	38	0·71
	Middle	0·42	63	0·141	38	0·63
	Basal	0·54	65	0·144	39	0·65
Significant difference within shoot types.		0·12	13	0·025	6	0·07

contains more than either of the earlier-formed ones, while with manganese the apical leaf contains more than the middle one but about the same amount as the basal one. In general these results fit into a chronological sequence, since the X type of shoot is generally beginning growth not much before the V type finishes growth, while the long shoots continue growth for some time after the secondary spurs have ceased. Thus the leaves formed early in the season have high contents of these four elements; those leaves formed in mid-season have lower values, while in the leaves formed at the end of the growing season the content of each element, except calcium, is again somewhat higher. The fact that the basal leaves of the long shoots have lower mineral contents than the leaves of the non-fruiting spur may be due to some remobilization from the former to supply the requirements of the younger leaves on the same shoots.

The distribution of potassium among the various types of leaf is rather different from that of the other elements. The highest content is found in the middle leaves of the long shoots and the next highest in their apical leaves; the leaves of the non-fruiting spurs have a uniformly low content of potassium, and those of the secondary spurs also have a low content, with a tendency to rise towards the apex. These results may well be associated with the exceptional mobility of potassium in the plant; some of the potassium of the older leaves may be translocated to the leaves formed at the end of the season.

It will be noted that the data on potassium distribution reported here are in agreement with the observations of other investigators (with the possible exception of Kidson *et al.*, 1940),

but that the data on magnesium content of different leaves on the long shoots are not. Comment on these disagreements would at present be premature.

Conclusions regarding the value of different types of leaf for analytical diagnosis of nutritional level can be drawn only in respect of those elements for which significant differences between trees have been demonstrated—i.e. potassium and manganese. As to potassium, there is a significant interaction between the effect of potash application and the leaf and shoot types, this being mainly ascribable to the exceptionally small difference between the means for the basal leaves of the long shoots. In Table III the figures are expressed on a relative basis, since the errors of sampling, and of analysis when a spectrographic method is used, are roughly proportional to the quantity present.

TABLE III.

Difference (as percentage of their mean) between potassium content of leaves of trees on plots with single and double potash applications.

Leaf position.	Shoot type.		
	X.	V.	L.
Basal	58	57	23
Middle	54	49	35
Apical	46	57	37

It will be seen that the proportional differences between the two treatments are greater in the spur leaves (X and V) than in those of the long shoots (L). The interaction of the difference in potassium content between the two ends of the plantation with the leaf and shoot types is not significant, except that the differences between basal leaves are greater than between middle leaves. But this interaction is highly significant with manganese. The greatest proportional differences in manganese content are found in the basal leaf of the non-fruiting spur, as is shown in Table IV ; the difference in the basal leaf of the long shoot is also large, but that in the apical leaf of the same type of shoot, which has often been sampled for analytical diagnosis, is relatively small.

TABLE IV.

Difference (as percentage of their mean) between manganese content of leaves from east and west ends of the plantation.

Leaf position.	Shoot type.		
	X.	V.	L.
Basal	54	88	81
Middle	79	66	73
Apical	71	70	57

The practical conclusions that may be drawn from these results in respect of assessing the nutritional level of an apple tree by leaf analysis are :

(i) that, since different morphological types or leaf differ considerably in their mineral content, it is important that comparisons between trees should be confined to samples of the same type ; and

(ii) that until further information becomes available the most promising type of leaf for this purpose is the basal leaf of the non-fruiting spur.

It is evident, however, that similar investigations need to be carried out on trees known to differ in nutritional level in respect of elements other than potassium and manganese, and

confirmation of the present results on trees showing a wider range of potassium and manganese nutrition is desirable. The experiments are already being extended to other times of the year, and to seasons with a less heavy crop than that of 1940.

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SUMMARY.

Samples were taken of apical, middle and basal leaves from primary long shoots, from primary non-bearing spurs, and from secondary spurs on bearing spurs of trees of Cox's Orange Pippin apple on Malling rootstock XII. The trees were receiving eight different manurial treatments. The samples were analysed for calcium, iron, magnesium, manganese and potassium.

Samples from plots receiving double potash applications contained 58 per cent. more potassium than those from plots receiving a single application; samples from plots receiving sulphate of ammonia contained 42 per cent. more manganese than those from the no-nitrogen plots.

The potassium content was highest in leaves from the middle and apex of the long shoots; the content of the other elements was highest in the basal leaves of the non-bearing spurs. In general, for all elements except potassium, the content declined as the date of leaf formation became later in the season, but rose slightly again at the end of the season.

Differences in manganese and potassium status of different trees are reflected more clearly in the composition of the basal leaves of the non-fruiting spur than in that of the other leaf types investigated.

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APPENDIX.

Mean mineral contents for each type of leaf on each tree.

Table V.

Calcium (percentage of dry matter).

Shoot type :		X.			V.			L.		
Leaf position :		Basal.	Middle.	Apical.	Basal.	Middle.	Apical.	Basal.	Middle.	Apical.
End of plantation.	Treatment.									
East	K ..	·50	·60	·56	1·56	1·31	·96	·60	·18	·16
	NK ..	·51	·46	·47	·88	·74	·86	·53	·16	·14
	PK ..	·32	·55	·36	·91	·42	·39	·51	·21	·12
	NPK ..	·34	·38	·44	1·36	·82	·56	·75	·16	·08
	2K ..	·62	·36	·53	·90	1·25	·61	·61	·34	·07
	N2K ..	·49	·22	·31	·54	·42	·67	·40	·28	·08
	P2K ..	·43	·58	·58	·98	1·07	·51	·84	·24	·19
	NP2K ..	·48	·43	·49	1·47	·92	·71	·62	·21	·03
West	K ..	·84	·34	·22	1·03	·56	·28	·41	·27	·03
	NK ..	·36	·33	·39	·98	1·04	·67	·44	·13	·23
	PK ..	·35	·32	·49	1·62	1·51	·63	·97	·19	·07
	NPK ..	·33	·50	·36	·56	·54	·52	·65	·12	·08
	2K ..	1·30	·46	·42	·98	1·45	·93	·77	·16	·24
	N2K ..	·76	·41	·48	1·41	·62	·42	·54	·31	·26
	P2K ..	·68	·38	·58	·36	·51	·57	·74	·25	·22
	NP2K ..	·41	·41	·49	·68	·40	·38	·52	·16	·11

Significant difference between leaves within shoots = 0·30.
Significant difference between shoots = 0·26.

Table VI.

Iron (p.p.m. of dry matter).

Shoot type :		X.			V.			L.		
Leaf position :		Basal.	Middle.	Apical.	Basal.	Middle.	Apical.	Basal.	Middle.	Apical.
End of plantation.	Treatment.									
East	K ..	57	70	76	121	119	97	73	72	44
	NK ..	44	62	75	81	50	52	56	62	114
	PK ..	69	64	49	81	42	54	61	94	108
	NPK ..	52	79	40	127	92	55	72	57	71
	2K ..	42	108	100	88	144	82	50	54	61
	N2K ..	80	66	57	68	71	85	64	90	113
	P2K ..	57	67	82	84	91	60	95	96	92
	NP2K ..	86	64	74	100	94	71	61	51	110
West	K ..	99	52	17	91	90	56	47	95	65
	NK ..	58	71	67	117	90	50	50	50	123
	PK ..	69	27	51	125	124	65	52	66	55
	NPK ..	59	45	74	90	66	47	65	37	71
	2K ..	92	68	56	98	120	88	50	38	84
	N2K ..	64	28	56	87	70	31	77	54	86
	P2K ..	70	66	77	52	55	91	115	76	96
	NP2K ..	49	69	58	58	87	34	76	125	117

Significant difference between leaves within shoots = 36.
Significant difference between shoots = 59.

TABLE VII.

Magnesium (percentage of dry matter).

Shoot type:		X.			V.			L.		
Leaf position:		Basal.	Middle.	Apical.	Basal.	Middle.	Apical.	Basal.	Middle.	Apical.
End of plantation.	Treatment.									
East	K ..	·182	·189	·119	·354	·275	·269	·207	·084	·066
	NK ..	·115	·135	·117	·280	·149	·123	·124	·050	·090
	PK ..	·138	·119	·109	·213	·124	·118	·151	·072	·070
	NPK ..	·110	·143	·070	·260	·229	·132	·204	·043	·053
	2K ..	·122	·162	·125	·204	·280	·142	·113	·060	·078
	N2K ..	·134	·109	·098	·156	·108	·112	·092	·079	·085
	P2K ..	·140	·152	·153	·311	·210	·172	·287	·065	·064
	NP2K ..	·107	·134	·135	·248	·202	·119	·154	·050	·056
West	K ..	·168	·149	·061	·252	·175	·083	·115	·065	·057
	NK ..	·136	·141	·139	·245	·246	·186	·120	·045	·133
	PK ..	·141	·102	·142	·353	·324	·220	·204	·063	·080
	NPK ..	·071	·080	·085	·143	·136	·107	·125	·037	·091
	2K ..	·239	·179	·171	·302	·314	·228	·165	·050	·095
	N2K ..	·187	·140	·120	·276	·166	·178	·182	·056	·141
	P2K ..	·237	·222	·176	·154	·153	·174	·279	·062	·080
	NP2K ..	·073	·110	·112	·149	·171	·077	·111	·050	·073

Significant difference between leaves within shoots = 0·066.
Significant difference between shoots = 0·120.

TABLE VIII.

Manganese (p.p.m. of dry matter).

Shoot type:		X.			V.			L.		
Leaf position:		Basal.	Middle.	Apical.	Basal.	Middle.	Apical.	Basal.	Middle.	Apical.
End of plantation.	Treatment.									
East	K ..	39	44	45	89	65	49	40	28	40
	NK ..	53	60	62	90	66	55	62	45	77
	PK ..	29	30	27	62	27	28	36	38	34
	NPK ..	46	73	55	110	92	66	123	47	91
	2K ..	43	61	57	66	80	61	28	24	34
	N2K ..	95	79	79	85	57	69	59	74	59
	P2K ..	35	30	31	57	56	44	59	43	38
	NP2K ..	52	49	60	119	96	68	74	55	70
West	K ..	30	19	12	27	27	20	16	14	25
	NK ..	25	23	24	43	37	30	20	16	33
	PK ..	22	16	19	38	37	21	25	20	24
	NPK ..	14	16	17	21	22	17	22	16	34
	2K ..	37	29	28	38	55	39	24	21	31
	N2K ..	47	33	39	54	38	34	38	27	39
	P2K ..	28	23	30	19	20	25	35	23	32
	NP2K ..	23	25	31	25	35	27	23	30	28

Significant difference between leaves within shoots = 18.
Significant difference between shoots = 29.

TABLE IX.

Potassium (percentage of dry matter).

Shoot type:		X.			V.			L.		
Leaf position:		Basal.	Middle.	Apical.	Basal.	Middle.	Apical.	Basal.	Middle.	Apical.
End of plantation.	Treatment.									
East	K ..	·40	·41	·54	·49	·47	·39	·50	·73	·63
	NK ..	·66	·57	·76	·67	·61	·59	·93	1·06	·79
	PK ..	·41	·42	·63	·62	·46	·49	·61	·75	·63
	NPK ..	·70	·69	·65	·55	·63	·65	·93	1·04	·97
	2K ..	1·00	1·01	1·11	·69	·84	·77	1·00	1·04	·95
	N2K ..	1·08	1·21	1·25	1·33	·96	1·25	·99	1·48	1·29
	P2K ..	·83	·72	·73	·88	·75	·78	·88	1·18	·97
	NP2K ..	1·12	·88	1·17	·88	·98	·93	·91	1·12	·99
West	K ..	·46	·44	·53	·39	·56	·54	·51	·82	·70
	NK ..	·46	·54	·64	·44	·43	·37	·72	·72	·90
	PK ..	·28	·27	·26	·22	·25	·27	·45	·62	·35
	NPK ..	·32	·36	·40	·32	·36	·34	·49	·71	·62
	2K ..	·85	·86	·87	·83	·91	·75	·63	1·18	1·15
	N2K ..	·52	·52	·63	·61	·64	·91	·71	·90	·87
	P2K ..	·62	·54	·58	·80	·54	·58	·77	·94	·85
	NP2K ..	·74	·71	·68	·64	·67	·81	·62	1·29	1·07

Significant difference between leaves within shoots = 0·18.
Significant difference between shoots = 0·38.

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PAPERY BARK CANKER OF FRUIT TREES IN RELATION TO SILVER LEAF DISEASE

By H. WORMALD
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In papery bark cankers of fruit trees the outer cell layers of the bark become separated from the underlying tissues to form a loose, thin, papery membrane. This condition arises when the cortical cells contain a superabundance of water. Certain cells in a layer a little way below the surface become swollen, thin-walled, bladder-like and full of sap. These cells collapse and, as their contents escape, turn brown and, in dry weather, form a powdery layer. In this way the continuity between the tissues on their outer and their inner sides is interrupted and the outer tissues thus set free form the papery bark. On young branches of cherry trees this condition has been referred to as " tan disease ", from the tan-like appearance of the dead, brown, powdery, cells which are exposed as the papery covering peels off.

Papery bark frequently occurs on apple trees after the branches have been cut back and topgrafted. The grafting is done in early spring when there is an abundance of sap in the living cells of the bark, and the surrounding atmosphere usually has a high moisture content. The removal of the main branches under these conditions is particularly favourable for the appearance of papery bark cankers. Such conditions also favour the entrance into the trees of parasitic fungi, particularly the Silver Leaf fungus *Stereum purpureum.*

Papery bark has been seen on pear trees also (Wormald and Harris, 1940), and the association of *Stereum purpureum* with papery bark has been noted on a double-worked pear stock (Wormald, 1935).

In September, 1935, the writer examined a number of Newton Wonder apple trees that had been topgrafted with Laxton's Superb in April of that year, and found that branches and stems of some of the trees were severely cankered. The cankers were for the most part elongated in the direction of growth, so that each showed two long almost parallel lines between which the outer layers were peeling off as papery bark (Plate I, Fig. 1). On some branches the scion shoots were dead or dying or bore silvery foliage.

Sections through the cortex underneath the areas where the bark was blistered but not yet peeled off showed swollen thin-walled cells in a region about three cells below the surface, the outer three-cell-deep layer of cork cells being pushed outwards as a thin membrane which would later have become papery bark. This blistering, the early stage of papery bark, is therefore of the nature of an intumescence.

The wood in the region of the cankers was discoloured, and isolations from this brown wood from four cankers yielded pure fungus cultures all similar in general appearance. This fungus was *Stereum purpureum*, for, when inoculated into plum trees (April, 1936) it induced silvering of the foliage within six weeks. In 1937 the trees were dead or dying, and in November some of them bore fructifications of *Stereum purpureum* ; control trees remained quite healthy.

In 1936 four more fungus isolations were made from the wood of papery bark cankers on apples. Three of these again yielded *Stereum purpureum*, and Plate I, Fig. 2, shows the result of inoculating a plum tree with one of these isolations. The fourth, however, was different in culture, and in an inoculation experiment it failed to infect plum trees. This fungus was kindly examined by Dr. W. P. K. Findlay of the Forest Products Research Laboratory, Princes Risborough, who identified it as *Polystictus versicolor*.

The grafted apple trees showing pronounced papery bark, and found by isolation and inoculation experiments to harbour *Stereum purpureum* in their wood, bore no fructifications of the fungus at the time they were examined in the orchard (middle of September). Dead or dying trees were lifted and some of the stems were piled together and left exposed on open ground with

the result that during the autumn numerous fructifications of *Stereum purpureum* appeared on the bark. Papery bark cankers may thus serve as ports of entry for the parasite causing Silver Leaf disease, and so favour its spread.

In 1940, apple trees were inoculated, using two isolates of *Stereum purpureum* from papery bark cankers, an isolate of *Stereum purpureum* from plum, and the strain of *Polystictus versicolor* isolated from the papery bark canker referred to above. Five young Lane's Prince Albert trees growing in pots were used, one for each fungal strain and one as control. About the middle of April five branches, half to one inch thick, were cut back on each tree; four of these were inoculated, the fifth being an extra control. The cut ends were inoculated from plate cultures, then covered with moist, sterile cotton wool, and cellophane. The control branches were inoculated with sterile agar.

By the end of June seven of the branches inoculated with *Stereum purpureum* from apple showed Silver Leaf. There was no silvering on the others and no papery bark appeared on any of the five trees. Silvering of the foliage was entirely absent from all the trees in 1941 and 1942. In March, 1943, the branches were sawn off and split so that the downward extension of discoloration in the wood from the cut ends could be measured; the results were as follows (the year of isolation is shown in brackets):

Stereum purpureum	from apple (1935):	7 to 8 cm.
,, ,,	from apple (1940):	4 to 35 cm. (average 24·5 cm.).
,, ,,	from plum (1934):	1 to 3 cm.
Polystictus versicolor	from apple (1936):	1 to 1·5 cm.
Control branches		0·5 to 1 cm.

Stereum purpureum was re-isolated from the wood of the tree showing most discoloration of the tissues, thus indicating that the fungus was still alive, although no silvering of the foliage had been noticeable during the previous two years.

That there was little discoloration with Stereum from plum may have been due to loss of virulence from continued growth in artificially prepared culture media. Most browning was obtained with the recent isolation from a papery bark canker. The length of discoloured wood in the tree inoculated with *Polystictus versicolor* was about the same as that of the controls.

That *Polystictus versicolor* has been isolated from the wood of a grafted apple tree showing papery bark, in this country, is worthy of note because in Australia and Tasmania this fungus is commonly associated with, and has been considered to be the cause of, the dying back of topgrafted fruit trees (Thomas, 1928; Ward, 1929). It was because of such failures from ordinary topworking that methods of frameworking were introduced—methods that have proved successful not only in rendering losses from grafting less frequent but also in bringing the worked trees more quickly into bearing again (Garner and Walker, 1938).

Polystictus versicolor is one of the commonest fungi on rotting stumps of forest trees and is generally considered to be purely saprophytic. That it can enter grafted trees is shown by its isolation from the wood of one of the apple trees examined, and by the early appearance of its fructifications on the dead cut-back branches after grafting, seen not only in Australia and Tasmania but also at times in Britain. Whether *Polystictus versicolor* may at times behave as a parasite is uncertain, but if it does have parasitic tendencies it apparently requires very favourable conditions (such as those inducing papery bark) for the infection to be harmful. In the experiments mentioned it failed to infect the wood of either plum or apple.

The writer has observed papery bark cankers that have shown no evidence of their association with *Stereum purpureum* (Wormald, 1939). Thus Plate I, Fig. 3, shows cankers induced by hard pruning in wet weather. On the other hand, the absence of papery bark from the inoculated and control apple trees in the experiment mentioned above is probably related to the restricted growth of the trees under pot conditions. It is concluded, therefore, that *Stereum purpureum* is not the cause of papery bark cankers on grafted apple trees but that the conditions favouring

papery bark are also those that favour infection by the Silver Leaf fungus, so that the two diseases frequently occur together. When this happens infection by Stereum is likely to be serious, and the affected trees may succumb.

From the results of a grafting trial, which included trees with papery bark cankers containing *Stereum purpureum* as described in this paper, Garner (1942) found that "Cankers [papery bark] were more numerous on topworked than on frameworked branches". Frameworking is thus recommended instead of topworking, for not only do the trees come into bearing again more quickly but there is also less chance of the occurrence of papery bark cankers and of Silver Leaf.

SUMMARY.

Topgrafting, involving cutting back large limbs of fruit trees, is often soon followed by the appearance of papery bark cankers on branches and stems. This is frequently accompanied by Silver Leaf.

The conditions favouring the formation of papery bark—cutting back the main branches when the tissues contain an abundance of sap—also favour infection by *Stereum purpureum.*

Isolations from the discoloured wood of a number of grafted trees showing papery bark all yielded *Stereum purpureum*, except one from which *Polystictus versicolor* was isolated.

The advantages of frameworking over topworking for apple trees are emphasized.

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(*Received* 28/5/43.)

PLATE I.

Fig. 1.

A papery bark canker, following grafting, on an apple branch.

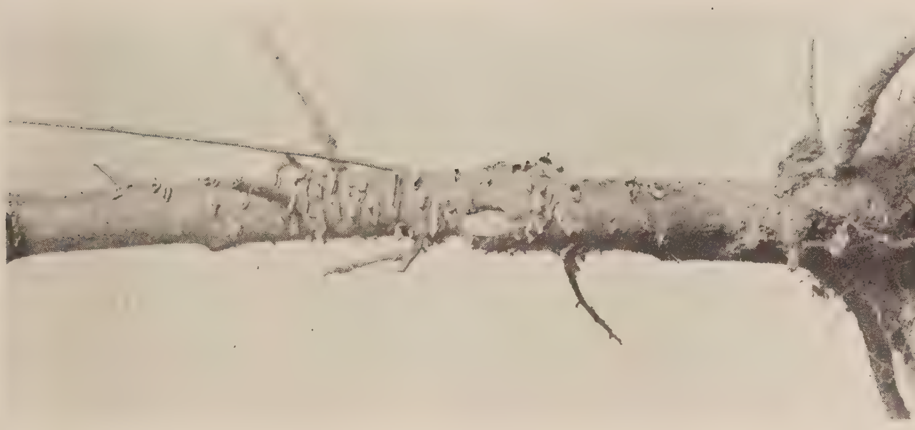

Fig. 2.

Stem of Victoria plum tree bearing fructifications of *Stereum purpureum*. The tree had been inoculated thirteen months previously with the fungus isolated from a papery bark canker on apple.

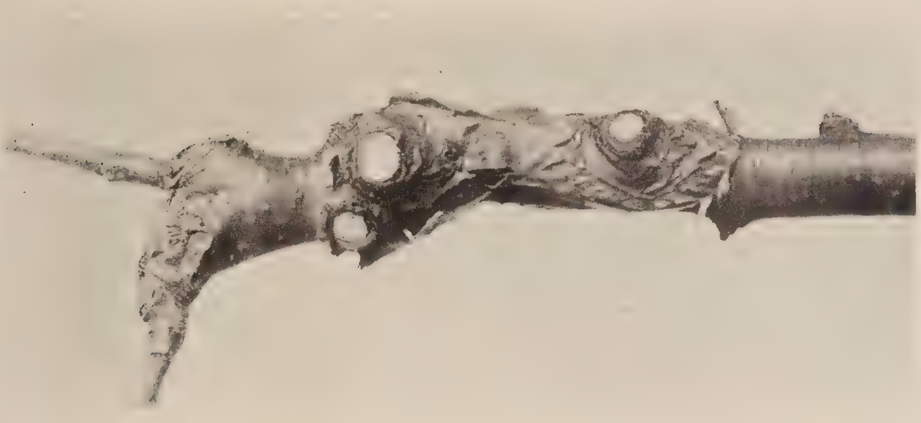

Fig. 3.

Papery bark cankers developing after hard pruning in wet weather.

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PLATE I.

Reconstructed root-system of Fuggle, Malling Soil Series.

In Plates I-III the photographs were taken from a height corresponding to the original ground level.

PLATE II.

Reconstructed root-system of Brewer's Favourite, Malling Soil Series.

Reconstructed root-system of Rodmersham Golding, Barming Soil Series.

PLATE III.

Reconstructed root-system of Fuggle, Barming Soil Series.

Reconstructed root-system of Fuggle, on poorly drained soil at Paddock Wood.

ROOT STUDIES

X. THE ROOT-SYSTEMS OF HOPS ON DIFFERENT SOIL TYPES

By F. H. BEARD
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MUCH valuable information on the root-systems of apple and other fruit trees and bushes has been obtained at East Malling (Rogers, 1932, 1933, 1934, 1935, 1939 a, b and c ; Rogers and Vyvyan, 1928, 1934). Similar knowledge of those of hop plants was badly needed in view of the recent outbreaks of the Verticillium Wilt disease which have appeared to be associated with certain soil types.

The usual practice of grubbing hops involves the removal—as completely as possible, to avoid the subsequent appearance of unwanted bines—of the crown, or rootstock, whose buds produce new bines that grow very rapidly and luxuriantly from May to July. Mere grubbing, however, is entirely insufficient to disclose the very extensive root-system which supports this growth ; detailed excavation is necessary for a thorough study of it. In the winter of 1938-39, therefore, such excavations were made of established hop plants of the varieties Fuggle, Rodmersham Golding and Brewer's Favourite, growing in two gardens at the Research Station and in one at Paddock Wood, on different soil types. It was hoped in this way to ascertain what influence soil type exerted on root development and whether and to what extent the variety of the hop concerned was also a factor in determining it.

The skeleton method of excavation, as described by Rogers and Vyvyan (1928), was used. As each root was uncovered its position and depth were recorded on a plan (Figs. 1, 2, 3) from which the root-system was afterwards reconstructed and photographed (Plates I-III). The spread of the roots is most clearly indicated by the plans, the depth and type of root by the photographs.

As the root-systems were found to be largely influenced by soil conditions, Mr. B. S. Furneaux of the South-Eastern Agricultural College very kindly made a thorough inspection on the sites ; and summaries of his report follow together with the observations on the root-systems made on each excavation.

I. ROOT DEVELOPMENT ON THE MALLING SOIL SERIES. (Church Field Garden.)

The soil is typical Malling loam, being a Pleistocene drift belonging to the Valley Gravels of the river Medway. The drift is very variable in depth and overlies the Ragstone of the Hythe Beds, so that drainage is very free.

(*a*) *Fuggle.*

Two seven-years-old plants were excavated here, one in the headland row and the other in the row adjacent. They had been planted as one-year rooted cuttings (sets) in October, 1931, at 7 ft. square.

The *Soil Profile* of the excavations was as follows : About 20 cm. of grey-brown medium loam, containing an appreciable quantity of sharp sand, passed into a warm brown, medium to light, loam, containing (besides sharp sand) some fine gravel made up of ironstone, flint and Ragstone fragments. Then came a slightly deeper coloured, rather sticky, medium to heavy, loam, containing rather coarser gravel. Finally came a darker, greyer, somewhat chocolate-brown, fairly compact silt, with a distinctly greasy feeling. This horizon is the " pug " layer, and is an accumulation of organic matter and clay immediately overlying the Hythe Beds

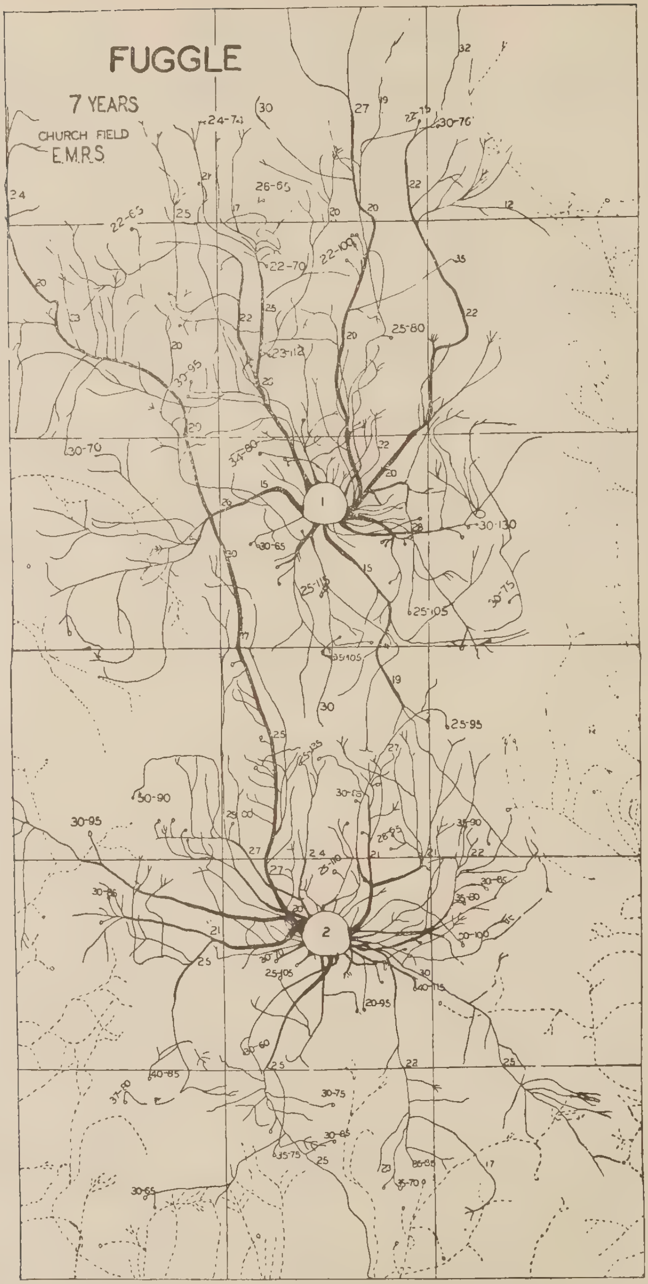

FIG. 1.

Root plan of two Fuggle plants, Malling Soil Series.

Numbers at the sides of roots indicate depth in cm. at these points. Small circles indicate the points at which the roots descended vertically; and the figures denote the upper and lower limits (in cm.) of the vertical portions. Broken lines indicate roots of adjacent plants. Heavier lines indicate principal roots, but are not drawn to scale. Each square = 1 sq. metre.

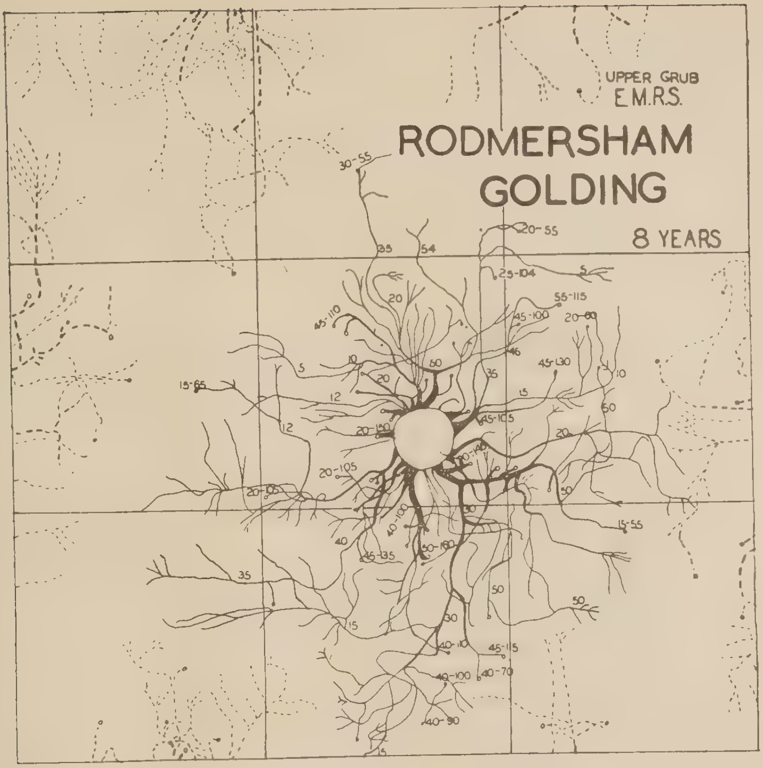

FIG. 2.
Root plan of Rodmersham Golding, Barming Soil Series.
For explanatory note see Fig. 1.

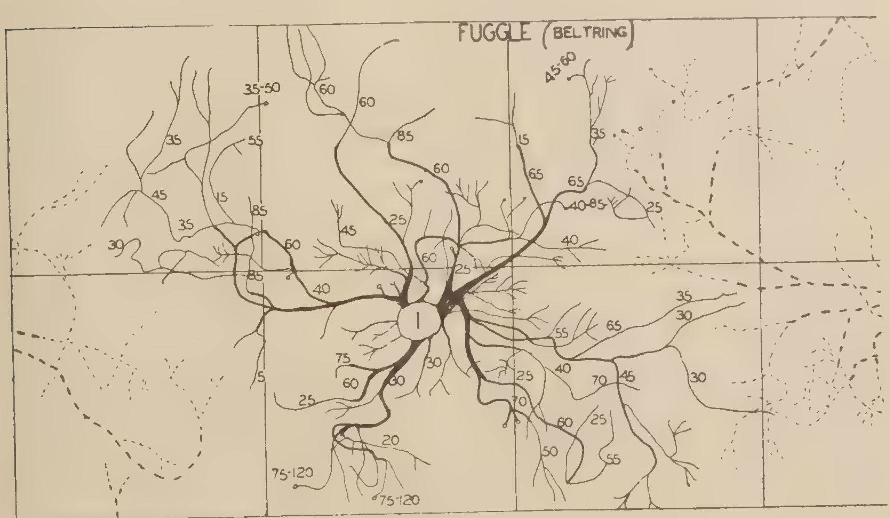

FIG. 3.
Root plan of Fuggle on poorly drained soil at Paddock Wood.
For explanatory note see Fig. 1.

(Bane and Gethin Jones, 1934). On the side of the excavation opposite to the profile described above, the soil was lighter throughout, being nowhere heavier than light loam, except for a very faint trace of " pug " immediately overlying the Ragstone, located at depths of from 65-116 cm.

The *Root-systems* of the two plants were practically identical. Both had a greater spread on the north than on the other sides, that of the headland plant particularly so. It will be seen from Fig. 1 that there was little interlacing of roots from adjacent plants. Rogers and Vyvyan (1934) found a similar condition in mature apple trees. An interesting exception was the large root, clearly shown on the left, which grew from the inner plant (No. 2), past the adjacent plant (No. 1), right out to the headland, as far as did many of the roots from the headland plant itself. It was about 4 metres long. No unsuberized roots were noticed (November.)

The root systems were found to consist of roots of two very clearly defined types:

(1) Main horizontal roots with many small laterals and much fibre, growing just below the cultivation level at a depth of from 20 to 30 cm. These roots were tough and wiry, and could readily be split but did not easily break. This is the type seen normally when established hops are grubbed in the Research Station gardens.

(2) From the main horizontal roots, and also from directly below the crown, medium sized roots descended vertically, many of them entering cracks in the Ragstone rock. These roots are clearly shown in Plate I. They were irregularly swollen, fleshy, readily broken and devoid of fibre. Branching rarely occurred, but when it did the branches usually descended in parallel. The bark was often split showing unsuberized areas. In extreme cases these swollen roots resembled those of a Brassica attacked by Club Root. This type of root has often been noticed on one-year rooted hop sets when lifted from the East Malling nursery, but it was not known that it persisted in mature hills.

As might be expected the two types of root have very different anatomical structures. The wiry horizontal type has a relatively narrow bark (i.e. all tissue outside the cambium), whilst the fleshy vertical roots have a very wide bark in proportion to the area of wood, about eight times as wide as in the horizontal roots of the same total diameter.

(*b*) *Brewer's Favourite.*

This is a hybrid raised by Professor Salmon of the South-Eastern Agricultural College, and is very different in character from ordinary European commercial hops. It ripens later than Fuggle (10-14 days later at East Malling) and normally makes a large amount of lateral growth after midsummer, when growth on Fuggle has practically ceased. The six-years-old plant excavated was one of a number planted in March, 1933, at 7 ft. square on ground adjoining the Fuggle plot slightly further up the gentle, northerly, slope.

The *Soil Profile* of the excavation was similar to that of the Fuggle but rather deeper, Ragstone being found at a depth of 110-130 cm.

The *Root-system* was similar in character to that of the Fuggle plants (see Plate II). It possessed, perhaps, a slightly higher, but insignificant, proportion of vertical roots, and was definitely less vigorous than that of the Fuggle plants, even after making allowance for the difference in age (one year). Possibly the strong lateral growth of the bine in the latter part of the growing season was supported by a considerable growth of fibre from the roots, which may perish at the end of the season and, in any case, would not readily be seen in a midwinter excavation. This idea receives support from the fact that Brewer's Favourite is known to be a very weak grower in the early part of the season. A few small unsuberized roots were found near the surface.

II. Root development on the Barming soil series. (Upper Grub.)

In this garden hops grow more strongly and more rankly than in the Church Field garden. A deep Pleistocene drift here overlies the Hythe Beds. At the surface the drift consists of a

shallow layer of brick-earth, below which is sand and fine gravel of the Valley Gravel group. The drainage is good but the sand is quite moist, and the faintly clouded colour suggests that it carries a small amount of spring water seeping through from the higher land to the south.

(*a*) *Rodmersham Golding.*

One plant of this variety was excavated from a number planted in 1930 at 6 ft. by 3 ft. to form layer beds for propagation purposes. After two seasons the layering was discontinued and the bines were trained up in the normal manner. In 1935 alternate hills in the rows were grubbed leaving the remainder at 6 ft. square.

The *Soil Profile* showed a top layer, of about 20 cm., of a greyish-brown medium loam, having a very smooth silky texture and being quite free from coarse sand ; this was followed by about 16 cm. of a warm brown, mellow textured, porous, medium loam of the brick-earth type ; below this came a yellowish-brown, loamy, sand containing a little fine gravel, the colour being clouded with browner and yellower shades. A little hassock (uncemented rock) was found at depths ranging from 115-130 cm., but no hard rock was encountered.

The *Root-system* was soon seen to be very different from that of Fuggle and Brewer's Favourite in the Church Field garden. No large roots were uncovered until the excavation had nearly reached the crown. Almost all the roots were of the vertical type, as is clearly shown in Plate II. The main roots turned steeply down, on leaving the crown, and branched after a short distance ; the relatively few horizontal roots and fibre were derived from upward growing laterals from the vertical roots. On the left of the photograph a root can be seen that has grown downwards for 50 cm., formed a complete U bend, and then grown upwards to the 20 cm. level, where it has given rise to a considerable amount of fine root and fibre. The crown and the large number of thick fleshy roots attached to it made an extremely heavy object. Some of the fleshy roots exhibited localized decayed portions from which, however, no pathogenic organism could be isolated. Such lesions may perhaps be a form of physiological breakdown due to the fleshy nature of the roots and possibly also to the moist condition of the soil. A few incompletely suberized roots were found. Traces of the large, dead, roots of the alternate plants grubbed in 1935 were plainly visible. There was no root competition.

(*b*) *Fuggle.*

The plant excavated was of uncertain age but probably arose from a runner remaining in the ground when some Fuggle plants were grubbed in 1934. It was, however, thought to be worth excavating, as the character of its root system would show whether the great differences in rooting between the Fuggle plants in the Church Field garden and the Rodmersham Golding plant in Upper Grub were due to soil conditions or to a varietal factor. The root-system of this Upper Grub Fuggle plant was, in fact, strikingly similar to that of the Rodmersham Golding in the same soil, as will be seen by comparing Plates II and III. Soil conditions, therefore, were responsible for the differences exhibited, not variety.

III. Root development of hops in poorly drained soil.

The soils of both the Church Field and Upper Grub gardens of the Research Station are, as already indicated, very well drained. For comparison, two Fuggle plants were excavated in a poorly drained garden at Paddock Wood, one in the better drained part of it, and the other in a very badly drained area. The ages of these plants were unknown. The hills were at 6 ft. square. The parent material of the soil is a Pleistocene drift, one of the Valley Gravel deposits laid down by the river Medway and its tributaries. The land is low lying, being rather less than 50 ft. above sea level, and is very badly drained in winter, having a water table which rises almost to the surface during wet periods.

The *Soil Profile* in the first excavation showed a top layer consisting of 23 cm. of grey-brown, heavy loam, free from stones ; this passed into a medium-brown, medium loam, faintly clouded

with orange-brown and bluish-grey shades. At 38 cm. there was light loam, clouded pale bluish-grey and orange-brown and containing a high proportion of very fine sand. At 61 cm. there was a light loam, rather more finely mottled, pale bluish-grey and dull, deep, orange-brown to brown. Occasional, small, rusty, almost black, iron oxide concretions (Crowstone) were present. At 89 cm. there was dark, grey-brown, iron-cemented, light loam, the sand in it being considerably coarser than in the horizons above. Some fine gravel, composed of sandstone fragments from the Hastings Beds (Lower Cretaceous), was present. From 125 cm. to the limit of the excavation (about 150 cm.) there was a light loam, mottled blue-grey and yellow-brown to orange-brown. The soil was porous throughout, except for the lower part of the top layer where a " pan ", about 10 cm. thick, resulting from repeated cultivation during wet conditions, was present.

In the second excavation the top layer was similar to that in the first ; at 23 cm. it passed into slightly stiffer pale, bluish-grey, silt, with orange-brown mottling and some darker grey clouding. At 35 cm. there was heavy silt, slightly deeper grey, with an increasing quantity of orange-brown (and also some black) iron oxide deposits ; these also increased with depth. At 56 cm. came very dark grey-brown (almost black) light silt, mottled brown and bluish-grey. At 74 cm. this passed into medium loam, slightly orange-brown flecked here and there with very pale, bluish-grey. Some fine grained sandstone gravel was present. From 99 cm. to the limit of the excavation (about 112 cm.) there was dark grey-brown gravel with some medium loam among the stones. The deposit of iron oxide was somewhat uneven ; in the western end of the pit the soil was cemented by it into numerous large, almost black, lumps, up to 8 cm. in diameter, while at the eastern end it was less plentiful and in the form of small concretions only. The pan, extending from 10-23 cm., was very compact and well marked. There was evidence that the water table sometimes reached to within 15 cm. of the surface, although at the time of the excavation (February 14th, 1939) it was at 94 cm.

The *Root-system* in the first excavation was rather weak (see Plate III). The main roots had grown rather crookedly down to about 50-60 cm. and then horizontally or even slightly upwards ; they were of a tough and wiry type. A few roots had grown down to the Crowstone, at 80-90 cm., and then horizontally for a short distance. Only two small roots had grown down into the Crowstone for any distance and these were blackened. As can be seen from the plan (Fig. 3), the roots, although rather weak and widely spaced, had reached out to touch those of adjacent plants.

The root-system in the second excavation was extremely weak, and the roots did not fully occupy all the available space. Its character was similar to that of the first plant, but the rooting was not so deep. Two fleshy, vertical, roots were noticed on one of the adjacent plants which might have been only recently planted.

IV. Discussion and conclusions.

As the excavation of mature hop plants is both laborious and expensive, it is not feasible to excavate as many as might be desired. However, the few excavated have shown that soil conditions are the primary factors influencing the type of root-system, and that any differences due to variety are so slight as to be obscured. On the other hand, Rogers and Vyvyan (1934) have shown that differences in the root systems of apple trees, due to rootstock influence, are clearly recognizable and persistent in spite of the powerful and general modifying influence of different soil conditions.

The Fuggle excavations showed that each of the selected soil types determined the type of the root system produced by the plant. On the well drained Malling loam of Church Field this consisted of main horizontal roots, with fibre, from which descended vertical fleshy roots growing down to, and even penetrating the crevices of, the Ragstone rock. The root-systems of hops growing on this soil, therefore, showed great similarity to those of apple trees in that soil (Rogers and Vyvyan, 1934). On the Upper Grub soil main horizontal roots were absent, the root-system

being characterized by a large number of fleshy, vertical, roots which descended either directly from the crown of the hill or within a very short distance of it. It is difficult to account for the absence of horizontal roots, but possibly a moist but well drained subsoil attracts the roots downwards.

On the poorly drained field at Paddock Wood the roots were characteristically twisted and bent and were almost entirely confined to a layer of soil about 35 cm. to 60 cm. deep. The hop root system was thus similar to that of apple trees on a heavy clay soil (Rogers and Vyvyan, 1934). There was evidence that the high water table limited the depth of rooting.

SUMMARY.

1. The results are given of excavations by the skeleton method of the root systems of a few mature Fuggle hop plants on three types of soil, and those of single plants of Brewer's Favourite and Rodmersham Golding on two of these types.

2. The soil profile and the root-system for each excavation is described.

3. Each type of soil—well-drained loam overlying porous rock, brick-earth over a moist but well drained loamy sand, and a poorly drained stiff loam with a high water table—is shown to induce the formation of a root system characteristic for the type.

4. Comparison is made between the hop root system and modifications in the root-systems of apple trees, which have previously been recorded, due to soil conditions.

5. Hop roots were found to be of two types: (1) Horizontal, tough and wiry, with considerable small root and fibre. (2) Vertical, fleshy and brittle, with little branching and no fibre. The proportion of each type of root was found to be governed by soil conditions.

6. Root plans and photographs of the reconstructed root-systems illustrate the text.

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The writer is greatly indebted to Dr. W. S. Rogers, for initiating him into the details of his excavation methods and for help and advice throughout the work; to Mr. B. S. Furneaux, for reports on the soil profiles at the several excavations; to Dr. H. Wormald, for examining the lesions on the roots of the hops growing on the Barming series; and to Miss E. C. Thompson for microscopical examination of the two types of roots. Thanks are also due to Mr. J. H. Waghorn, Manager of Messrs. Whitbread's Hop Farm, for permission to excavate plants on their Beltring farm; and to Miss K. E. Cornford, for photographing the root plans and reconstructed root-systems of which it is possible to reproduce only a few.

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BOOK REVIEWS

DECIDUOUS ORCHARDS. By W. H. CHANDLER. (London, Henry Kimpton, 1942, pp. 438, 109 figs., 21s. net.)

Students of pomology have been fortunate during the last twelve months in the appearance of two really first-class books on the scientific treatment of the subject : first, Gourlay and Howlett's *Modern Fruit Production*, and now Professor Chandler's work. Both approach the subject from the standpoint of the physiological responses of the tree ; both are equally wide in their scope. While the first, however, dealt primarily with American work, though including many references to British and other publications, the present work is even more general in outlook. The author states that " references have not been given to horticultural papers from which data for the text have been taken. Any writer, however, can be certain that if his paper appeared in the *Proceedings of the American Society for Horticultural Science* or the *Journal of Pomology and Horticultural Science* or was reviewed in *Horticultural Abstracts*, it has entered into what is said in this text or has prevented something from being said erroneously ". This can surely never have been truthfully said in such a publication before. The result is a most excellently balanced review of modern scientific knowledge on deciduous tree fruits and one which every student, research worker and up-to-date grower should read.

The first half of the book is devoted to the general physiology of tree fruits, beginning with processes in the setting and development of fruits and the processes of mature fruits, and passing on to orchard influences as they affect these. The second half is given up to special consideration of the different kinds of fruits. The print and general format are admirable and the sub-headings are brought out clearly in black type, making reference to any section much easier than is usual. The index, important in a book of this kind, is almost as good as the Index to *Horticultural Abstracts :* there can be no higher praise.

R.H.S.

THE DESIGN OF EXPERIMENTS. By R. A. FISHER, Sc.D., F.R.S. (London, Oliver & Boyd Ltd., Third Edition, 1942.)

The use of statistical methods for interpreting the results of experiments has become common in all branches of research work. Too often, however, even now, insufficient attention is paid to the original design of the experiments themselves. In the preface to the first edition of this book, the author states : " A clear grasp of simple and standardized statistical procedures will, as the reader may satisfy himself, go far to elucidate the principles of experimentation ; but these procedures are themselves only the means to a more important end. Their part is to satisfy the requirements of sound and intelligible experimental design, and to supply the machinery for unambiguous interpretation."

Despite the multiple pre-occupations of the times, scientific research is proceeding undeterred ; indeed, with new and urgent problems to solve. Intelligently designed experiments are economical in labour and save time, and it is encouraging, though hardly surprising, to find that there is sufficient demand for this " experimenter's textbook " to warrant a wartime edition.

Agricultural research has not been behindhand in adopting new and improved methods of experimental design, and many of the methods described in this book had their first test in agricultural experiments. But, in recent years, there has been a growing demand for better experiments in many other fields of work, and the principles upon which the design of experiments should be based, so ably set forth in the opening chapters of this book, apply equally well to all fields.

In laying out horticultural experiments on a considerable scale with perennial plants, it is usually wise to start simply ; for, during the long life of, say, an apple orchard, it may be necessary to add first one set of treatments and, later, others, as occasion arises. Thus, even though

he may not be able to foresee what these treatments will be, the designer should plan his experimental orchard to allow as far as possible for their inclusion. For this purpose the study of the more complex designs and the principles of confounding is likely to be well repaid. In this, the third edition of Professor Fisher's book, this subject has been further discussed, while the valuable contents of the previous editions have been retained. The book, therefore, will be more useful to the experimenter than ever.

T.N.H.

THE DIAGNOSIS OF MINERAL DEFICIENCIES IN PLANTS, BY VISUAL SYMPTOMS: A COLOUR ATLAS AND GUIDE. By T. WALLACE. (London, H.M. Stationery Office, 1943, pp. 116+vi, 114 col. pls., 10s. net.)

Realization of the extent to which deficiencies of the so-called "minor" or "trace" elements may cause depression of crop-yields has, during the last two decades, stimulated a vast amount of research in all countries into the roles of these elements in plant nutrition, the nature of the injuries produced by such deficiencies and methods for their detection and control. At first it seemed a relatively simple problem to determine, by sand- or water-culture, the characteristic symptoms produced by lack of any particular element, to recognize these symptoms in the field and to cure the trouble by supplying the necessary element to the soil. Further, it seemed that by soil analysis, deficiencies of any elements could be determined and immediately corrected.

Soon, however, it became apparent that matters were much more complex than this, that different plants showed different symptoms with the same deficiency and that the effects produced on the plant depended not merely on the actual measurable concentration of any one element in the soil solution, but on the relation of that concentration to the concentration of other elements. To illustrate this latter point one need only instance the case of lime-induced chlorosis, where iron-deficiency symptoms occur in plants on chalky soils even when these soils contain relatively abundant iron.

The first step, however, must necessarily be the recognition of the characteristic symptoms of a deficiency in a particular crop. Hitherto, anyone unfamiliar with the symptoms has had to rely on printed descriptions of deficiency symptoms, or to search the literature for the scattered coloured illustrations published in scientific journals, mostly American. It is true that Hambidge's *Hunger Signs in Crops* was published in America in 1941, but this has not been readily available in this country; and, in any case, the colours in it are so variable in different plates that in some cases the healthy plant of one series closely resembles the deficient plant in another.

In this new publication, Dr. Wallace has brought together a most admirable series of colour plates illustrating a wide range of deficiency symptoms of the more important horticultural and agricultural crops. Since all the plates were taken by one photographer, with one colour-plate process, there is a most satisfactory uniformity of colour reproduction which makes the whole series a true "Colour Atlas and Guide".

In a few concise and admirable chapters the author discusses the essential points of plant nutrition, soils in relation to nutrient supply, methods of determining mineral deficiencies and the recognition of deficiency symptoms.

Dr. Wallace has produced a volume of first rate importance not only to research workers and advisory officers but to all growers, every one of whom would benefit by carrying a copy in his pocket when walking his fields. The Agricultural Research Council, under whose ægis this work was produced, is to be congratulated on making possible the appearance of such an invaluable guide under wartime conditions.

R.H.S.

THE JOURNAL OF POMOLOGY AND HORTICULTURAL SCIENCE

INDEX TO VOLUMES XI–XX

Compiled by E. E. Cheesman,
East Malling Research Station, Maidstone, Kent.

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The Publication Committee would like to express their indebtedness to Dr. E. E. Cheesman, who undertook the compilation of this Index. Like that for the first ten volumes, it will be of great value both to research workers and the readers of the *Journal* generally.

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INTRODUCTORY NOTES

Index of Contents

This index presents exact titles and authorship with all the data necessary for citation of articles appearing in the *Journal*. It should be used for checking titles and authorships, which are often abridged or otherwise altered in the Subject Index and Author Index.

Subject Index

The plan here is similar to that of the index to the first ten volumes of *The Journal of Pomology and Horticultural Science*, which was issued in 1933, titles being indexed under as many headings as convenient.

Entries in **bold-face type** followed by an author's surname represent titles of articles, though these are often modified or re-arranged to suit the alphabetical listing and preliminaries such as The, Notes on, Studies of, are sometimes omitted. The exact title and authorship, when required for citation, can be quickly obtained from the volume and page numbers by turning back to the Index of Contents.

The other entries are subject references under heads which may or may not appear in the title. No attempt has been made to index every occurrence of names of plant varieties, insects, fungi or chemical substances mentioned in the text. Varietal names as a rule are indexed only where they either occur in a title or are coupled with descriptive notes in the text. Where lists of names occur in papers they are indicated under a general heading: for example, a list of cherry varieties observed with reference to their susceptibility to bacteriosis, occurring on p. 29 of Volume XV is indexed under *Cherry varieties, susceptibility to bacteriosis* as XV 29 (list) and the hundred or so varietal names do not themselves appear in the index.

Pests and diseases dealt with at any length are indexed under their scientific names, under their ordinary English names and under *Pests* and *Diseases* as well as under their hosts. Chemical substances not indexed directly may be traced through general headings such as *Growth substances*, *Manures* and *Sprays*.

Author Index

The author index is arranged in alphabetical order of authors' names, and in chronological order of articles contributed by individual authors. Articles having more than one author will be found indexed under the name of each author, only one name being shown in full for each entry. Where reference is being made for citation, therefore, authorship should be checked by referring back to the Index of Contents. This author index includes only names of authors of articles appearing in full in the *Journal*, and not those of authors of books reviewed. The latter will be found under "Reviews of Books" in the Subject Index. Authorship of reviews is not indexed.

E.E.C.

DATES OF PUBLICATION

Vol.	Number	Date	Pages
Vol. XI.	1	March 1933	1- 80
	2	June 1933	81-176
	3	September 1933	177-258
	4	December 1933	259-334
Vol. XII.	1	March 1934	1- 80
	2	July 1934	81-176
	3	October 1934	177-248
	4	December 1934	249-328
Vol. XIII.	1	March 1935	1- 80
	2	June 1935	81-148
	3	October 1935	149-260
	4	December 1935	261-360
Vol. XIV.	1	April 1936	1- 96
	2	July 1936	97-204
	3	October 1936	205-298
	4	January 1937	299-394
Vol. XV.	1	April 1937	1- 68
	2	July 1937	69-164
	3	October 1937	165-252
	4	January 1938	253-356

Vol.	Number	Date	Pages
Vol. XVI.	1	March 1938	1-100
	2	June 1938	101-184
	3	September 1938	185-290
	4	January 1939	291-402
Vol. XVII.	1	March 1939	1- 84
	2	June 1939	85-184
	3	September 1939	185-292
	4	January 1940	293-390
Vol. XVIII.	1	March 1940	1- 88
	2	June 1940	89-176
	3	October 1940	177-296
	4	January 1941	297-394
Vol. XIX.	1, 2	August 1941	1-148
	3, 4	March 1942	149-276
Vol. XX.	1, 2	August 1942	1- 68
	3, 4	October 1943	69-156

INDEX OF CONTENTS

VOLUMES XI-XX

VOLUME XI

VOLUME XII

VOLUME XIII

VOLUME XIV

VOLUME XIV. No. 4. JANUARY 1937.

VOLUME XV

VOLUME XV. No. 1. APRIL 1937.

VOLUME XV. No. 2. JULY 1937.

VOLUME XV. No. 3. OCTOBER 1937.

VOLUME XV. No. 4. JANUARY 1938.

VOLUME XVI

VOLUME XVII

VOLUME XVIII

VOLUME XIX

VOLUME XX

SUBJECT INDEX

VOLUMES XI - XX

AUTHOR INDEX

VOLUMES XI - XX

67